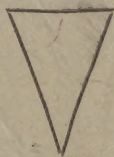


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# SYLLABUS OF MALARIOLOGY



Modified and Re-Edited

January 1946

for the

ARMY SCHOOL OF MALARIOLOGY



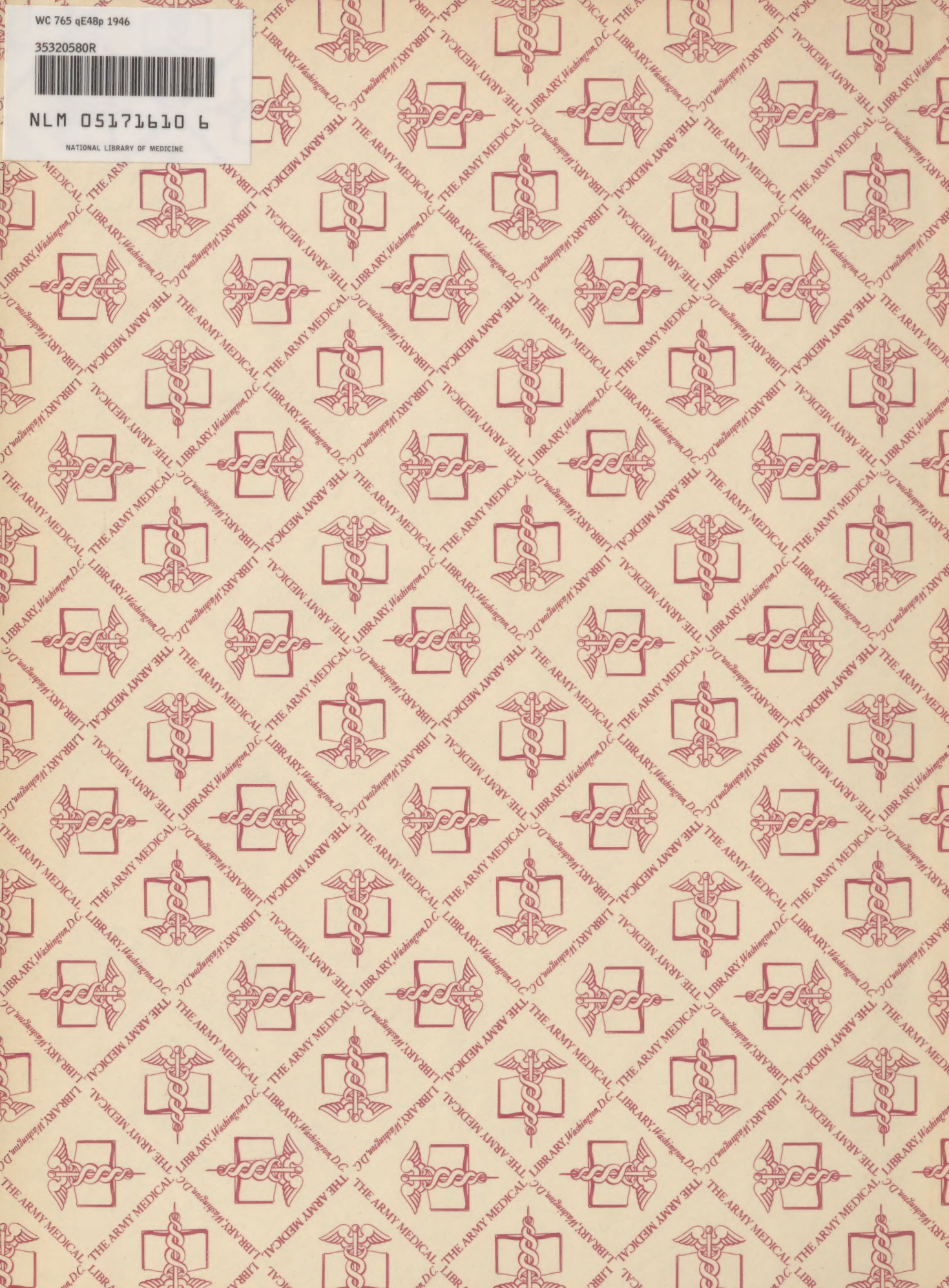
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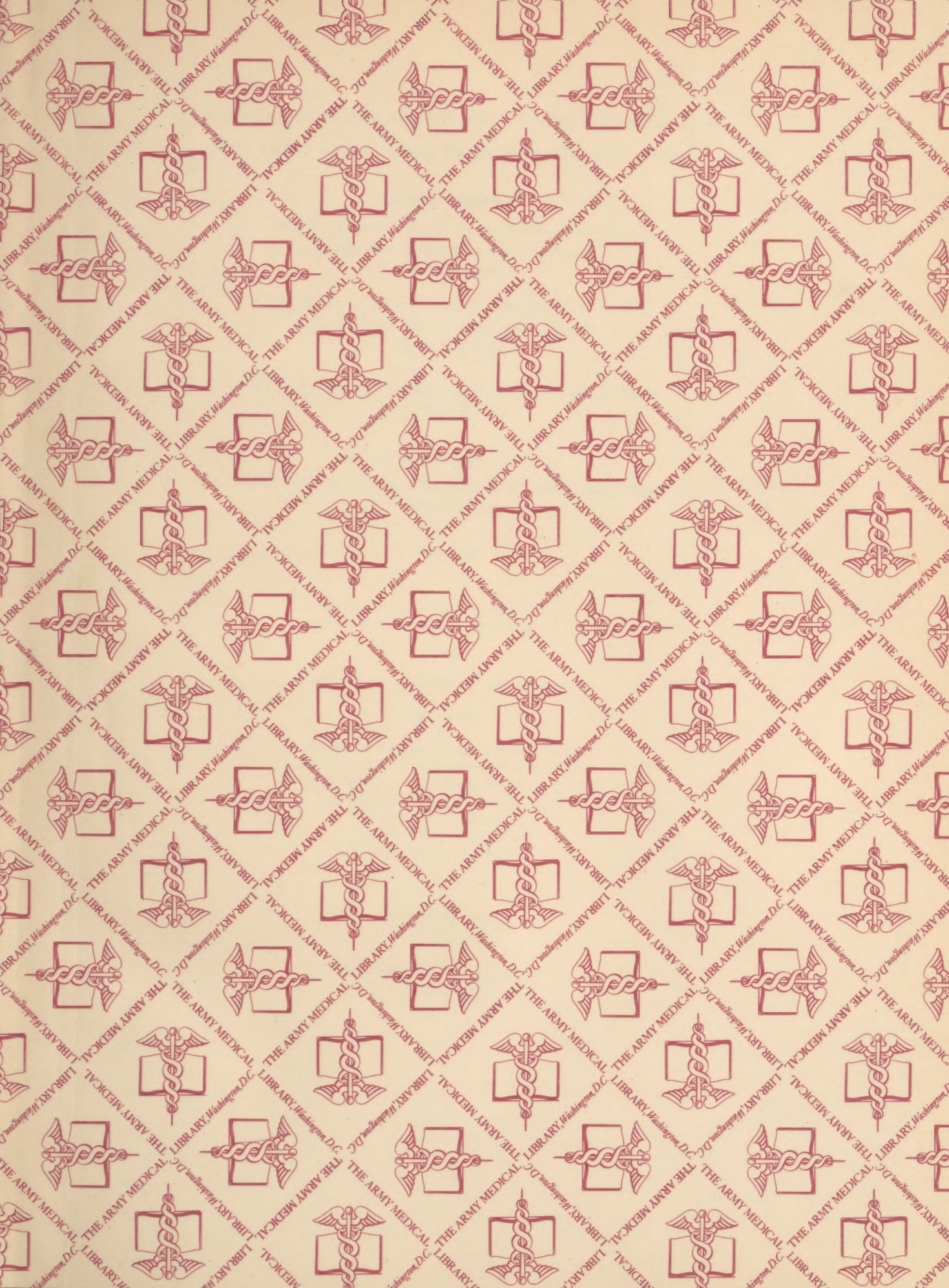
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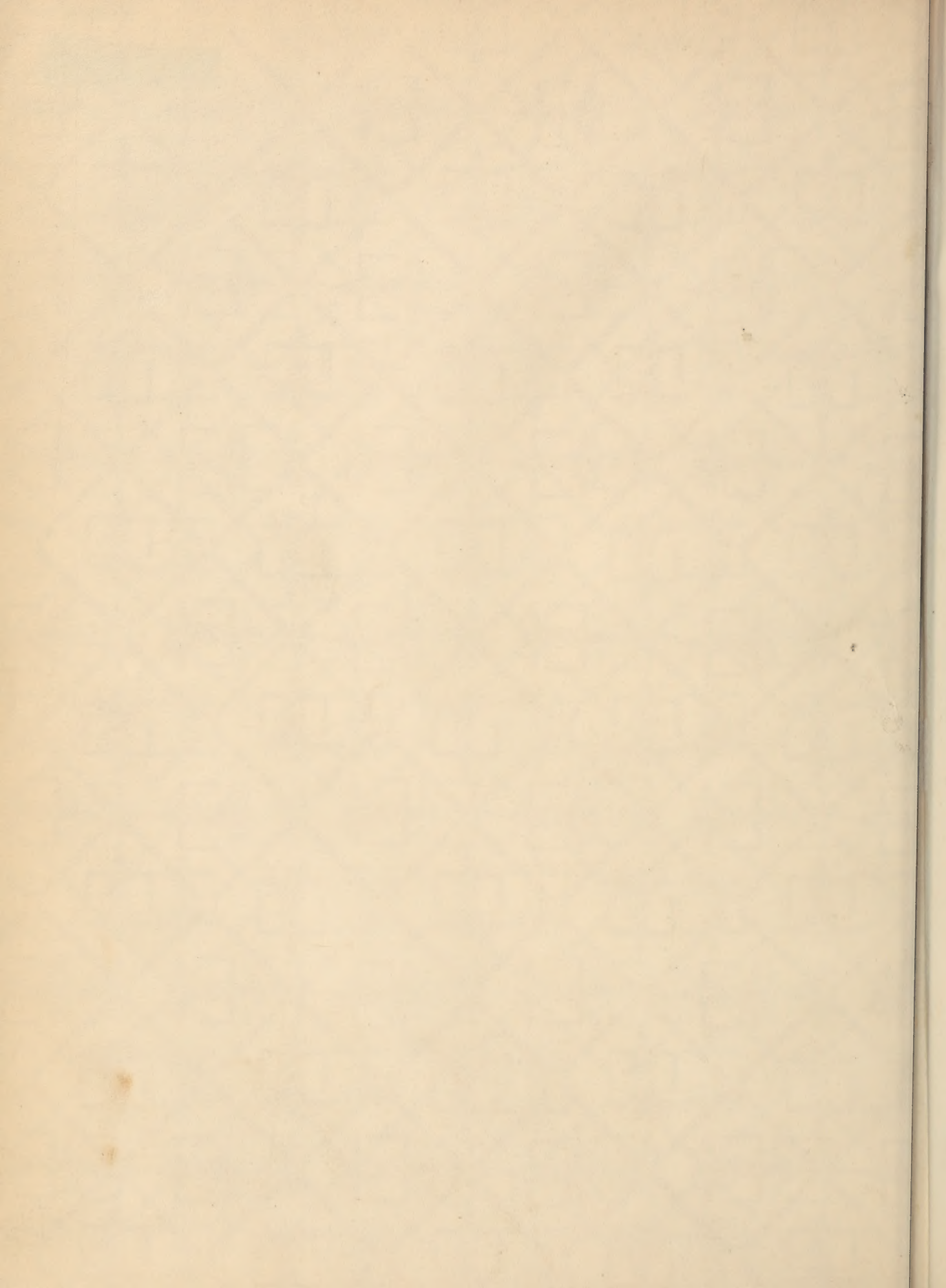


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**PRACTICAL MALARIA SURVEY AND CONTROL PROCEDURES  
AND THEIR APPLICATION TO EMERGENCY SITUATIONS**

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Including

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and

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## FOREWORD

The advent of World War II presented many problems of mass production. Included in this was mass training for special assignments. Never before had a pressing emergency demand existed for the protection of so many individuals from the disease malaria in so many varied localities of the world. An adequate supply of trained personnel did not exist to supply this need. During the period of rapid training of military personnel in malaria control procedures, it became evident that a single compact field reference manual was indicated, presenting comprehensive developments of malaria investigation and control, and outlining procedures, as modified by their application to an emergency situation.

The present syllabus was originally prepared for the malaria training course for military personnel which was conducted under the auspices of the Florida State Board of Health and subsequently, on two occasions, modified and re-edited in accordance with new information, has been dispersed to indicated military personnel by the Army School of Malariology.

JOHN E. ELMENDORF, JR.,  
Colonel, Medical Corps,  
Commandant of the  
Army School of Malariology

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## PREFACE

The following outline was prepared for a special purpose; namely, to acquaint physicians, entomologists, and engineers, recently assuming their new duties, with some of the fundamental facts of malaria investigation and control, and further to serve as a ready and convenient reference, a "Vade mecum", when in the field.

From the circumstances associated with the assumption of their new duties, many so assigned did not have the opportunity for long experience in activities of malaria investigation and control. The concepts expressed in this outline aim to be fundamental and basic. However, it should be borne in mind by those consulting this syllabus that the general practice of medicine is, as yet, not entirely a science and malaria prevention offers no exception to that statement. Scientific application of preventive measures is necessarily limited by existent scientific knowledge. Accordingly, in many phases of malaria study and control operations, information and conclusions cannot be presented in a dogmatic manner. In the final analysis, the individual personal test of common sense, logical thought, practical use, field experience, all associated with an open mind for evaluation of new procedures, should be applied before accepting or rejecting dictums expounded for the study and control of the disease.

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## GENERAL CONCEPTS OF MALARIOLOGY

The investigation of malaria naturally falls into three categories, the clinical, the entomological, and the engineering. Control involves a close co-ordination of applicatory procedures in these three divisions.

Clinical studies outline and define the problem, locate the disease geographically, and, through the analysis of the material secured, define the phase in which the disease exists as well as its intensity. Entomological studies determine what may be defined as the "why" of malaria. Having located, by the clinical procedures, the presence of the disease geographically, it is evident that vectors must have been within flight range of these cases at some time, unless the infected may have been imported from other malarious districts. Axiomatically, it may be stated that the presence of cases of malaria indicates a vector breeding in the locality, and conversely, the presence of breeding of a vector presages actual or potential malaria.

When malaria exists in an area, it exists because nature, frequently aided and abetted by man's activities, has decreed it, and has provided the essential factors involved in the propagation of the disease. The relationship of breeding areas to a blood meal is not a chance occurrence. Breeding occurs and persists in suitable water areas because of the accessibility of the blood meal, and because meteorological conditions, favorable for the propagation, exist as well. If these conditions favor the prolongation of the vector's life for sufficient time to permit of the development of the parasite, and its injection into humans, the disease malaria can result.

The female anopheline, in transmitting the disease, is performing simply an essential function of her own life, propagation of her own species. For that function she needs blood meals and suitable water areas. Malaria is an unfortunate by-product of the life history of those female anophelines, which, in search of a blood meal, bite man.

Many anophelines, actual or potential vectors, secure their blood meals from sources other than man and, accordingly, do not transmit the disease.

From the standpoint of life processes, it should be remembered that malaria is a natural phenomenon, though unfortunate for the individual or the localities suffering from it. To combat it effectively, knowledge of the natural factors involved must be complete. The malaria survey supplies the information upon which such an understanding is based. It is obvious that comprehensive plans for permanent malaria control demand studies of orientation in all phases of the subject. This is accomplished by execution of the classical malaria survey. With the present day knowledge of control procedures, the subject of the survey cannot be dismissed by the

characterization that it is academic. The first steps in the control of the disease malaria are a complete understanding of all procedures, and acquisition and analysis of essential facts. Application of the indicated procedures, revealed by the survey, to the practical field conditions found will then follow logically.

Certain procedures of malaria control activities, directed toward a curtailment of incidence of clinical manifestation of the disease, do not demand detailed pre-studies before their application to a field problem is justified. Under conditions encountered in war time, with bodies of troops moving rapidly from place to place through malarious districts and remaining for only short periods of time in such districts, there are certain procedures which can be applied on the basis of the simple knowledge that malaria exists or has been known to exist in the district. These measures involve those control activities included in large measure under the heading "Personally Applied Measures of Control". They comprise such activities as use of repellent, protective clothing, mosquito nets, head nets, and gloves, aerosol adult killing solutions, taking of suppressive medication to curtail manifestations of the disease, and the one procedure, classified as environmental control, mosquito proofing.

When malaria control activities, with the exception of mosquito proofing, are directed toward environmental factors for control of the disease, then, in the light of our present day knowledge, facts must be known.

To eliminate the vector in its aquatic stage, the vector must first be identified and breeding localities determined, irrespective of what means of elimination of the vector be employed; whether it be by larvicides, draining, filling or special management of water breeding areas.

To understand the magnitude of the problem involved, the status of the disease, be it epidemic or mild, moderate, severe, or hyperendemic, must be defined. To utilize natural means of control to the highest degree of efficiency, the ecology of the vector must be perfectly known. To foretell the dangers inherent in the infection existent in any locality, the presence or absence of the parasite, P. falciparum, must be determined. To utilize, to the best advantage, seasonal changes in the activity of the disease, such seasonal manifestations must be investigated. To establish control over parasite and gametocyte carriers, seed bed of the infecting agent, blood smears must be examined and cases with parasitemia identified.

The application of personally applied measures of control for emergency situations demands that malaria education of the personnel involved be complete and malaria discipline be developed and established, that all materials and equipment be used correctly and thoroughly under the immediate supervision of the malaria control personnel.

If more simplified means of control be demonstrated as practical, then, and only then, may the survey come to be regarded as academic.

PRACTICAL APPLICATION OF MALARIA INVESTIGATION  
AND CONTROL PROCEDURES TO AN EMERGENCY SITUATION

1. GENERAL ASPECTS OF MALARIA CONTROL

The procedure of an orthodox malaria survey is discussed with considerable detail elsewhere. (See page 35). The present discussion will treat of the subject of the practical application of survey and control procedures to an emergency situation.

In permanent communities, when dealing with the permanent control of malaria during peace time, the procedures used in both survey and control will differ from those which forcibly must be used in situations of an emergency nature.

The factors necessary for the propagation of malaria are the same under all conditions, and include the following:

The presence of suitable vectors (anophelines) in suitable densities.

Reservoirs of plasmodia in the blood of humans in the correct phase of development of that parasite, or the presence of the parasite in the vector (acquired from a previous human case) in stage of development capable of causing infection.

Suitable numbers of persons susceptible to the disease (non-immunes).

Access of infected vectors to the non-immunes.

The detailed study of the various factors, mentioned above, constitutes the essence of a malaria survey and reveals the points of attack feasible for instituting methods of control.

The propagation of malaria can be reduced to a simple concept: A human reservoir of parasites, and anopheline vector to transmit, and non-immunes. The mere mention of these factors suggests various manners of combating and controlling the disease.

General procedures for malaria control are frequently classified in accordance with various concepts of measures employed in such control, but here are classified in accordance with the link of the chain of the malaria cycle (man and vector and the associated parasites) toward which the attack is directed.

a. CONTROL THROUGH ATTACK ON VECTOR

(1) The aquatic stage

The vector can be attacked in its larval stage or as an adult. The

most classical permanent method recommended, to the present, is directed against the larval stage, and consists of drainage of those waters on which the eggs are deposited, and which are, at the same time, suitable for the development of eggs through the larval and pupal stages to the stage of adults. This constitutes real prevention and prevents the development of the disease at one of its principal sources. No water areas for breeding purposes, no Anopheles -- no Anopheles, no malaria. That is theoretically possible, but sometimes practical issues prevent an easy realization of this end.

Fill of areas holding water, and capable of breeding the vectors, is another most permanent method of elimination.

Another method directed against the larval stage is larvicides, principal among these being oil and Paris green\*. Their function is simple: Oil is reputed to prevent the larvae from projecting their syphons or breathing tubes through the surface for breathing, and they suffocate. Probably more important is the fact that many oils, in addition, possess toxic qualities, which kill the larvae. Paris green is ingested, as the larvae feed, and they are poisoned. It should be remembered that correct quantities of Paris green used for larvicidal purposes will never poison animals.

There are varying types of larvicides, and it should be borne in mind that frequently chemical wastes, present in the locality, may be utilized. In the absence of oil or Paris green, any such waste, which appears likely to be lethal, can be tested in a receptacle of water containing larvae, and the lethal effect on larvae noted.

In association with certain enterprises and activities, there may be available used oil from machines such as cars, trucks, airplanes, motorcycles, ships, etc. This oil, though not representing the preference of choice, can be utilized, especially when diluted with a grade of thinner oil. Heavy oil will not spread as readily over the surface of the water as special larviciding oils, and more care will be necessary in its application in order to realize a complete coverage. However, if applied correctly, it will function in a reasonably satisfactory manner. Quick and efficient kill can frequently be effected by application of a thin oil; whereas, killing of surface vegetation, and more lasting larvicidal effects, will result from a simultaneous or subsequent use of heavy oil.

Directed against the larval stage, predators can be employed, the most important of these being the top-feeding minnows.

Also, at times, salt water can be admitted to the fresh water pools, and a resulting concentration of salt developed to the point where anophelines cannot breed.

\*Note: The substance commonly known as DDT is discussed on pages 69-82.

Other methods directed against the larval stage and utilizable under certain conditions of limited application are such procedures as flooding, fluctuation of water levels, influencing the degree of sunshine by planting or felling trees, sluicing, or flushing, etc. These procedures are usually indicated because of specific circumstances, and accordingly, their application, as a general solution to the problem, is correspondingly limited.

With the exception of complete drainage or fill, no method directed against larvae will result in all instances 100% perfect. In fact, the results will fall far short of 100% perfection. The effectiveness of the applications directed against larvae will depend upon the resulting closeness of contact between the larvicide used and the larvae. Vegetation prevents oil, and also Paris green to a degree, from spreading over all portions of the surface of the water, and coming in sufficiently close contact with larvae to realize a kill. In like manner, fish are impeded from their attack on larvae by vegetation and debris. Currents, or lack of currents, in the water area may influence the distribution of substances used as a larvicide, and accordingly, detract from their value as larvicidal agents.

Water areas have to be larvicided constantly during the breeding season. The interval between application depends upon the period of time necessary for the breeding to progress from the egg stage to the adult. This period will differ in accordance with factors of rain, temperature, sunshine, available food, etc., and must be studied for each locality, but it is rarely found to be necessary at less than a weekly cycle. It must be remembered that Paris green does not kill eggs, very young larvae, nor pupae, and, accordingly, the time interval between applications is influenced by these facts.

## (2) The adult stage

The adult mosquito can be attacked in several manners to the end that she does not serve as vector and function, between the sick and the well, as the link, or carrier, of the parasite.

Mosquito proofing of houses and the use of mosquito nets play a dual role in preventing the mosquito from infecting men, and in preventing uninfected anophelines from acquiring infection from infected men. The value of these procedures should never be overlooked. They can be most effective.

Spray-killing of adults with various sprays has demonstrated its worth in the hands of various observers. In India it functioned effectively against Anopheles culicifacies, and reduced malaria rates drastically, as measured by splenic and blood findings.

Killing of all mosquitoes in habitations every morning, by any method at hand, even the use of the swatter, has proven to have distinct value in reducing the incidence of the disease.

Capitalizing upon an additional source of blood meal, when accidentally finding one or even making such a source of blood meal available, can function as a diversionary measure to prevent anopheline vectors from biting man and so transmitting the disease.

#### b. CONTROL THROUGH ACTIVITIES DIRECTED TOWARD MAN

Man, the carrier or reservoir of infection, can be treated with medications with the aim in view of preventing symptoms, or curing the disease, and, also to sterilize him as a carrier of malaria parasites; so, preventing the infection of other anophelines.

(NOTE: It should be remembered that, up until the present, there is no causal prophylactic against the organism (sporozoite) introduced with the bite of an infected mosquito. No known medicine will kill the sporozoite once it is injected into the human body by the mosquito bite. Continual suppressive medication can, and most frequently does, limit an infection to subclinical manifestations, and permits the individual to continue in the performance of his normal duties. If medication be withdrawn, clinical symptoms frequently appear without the necessity of a reinfection).

Man, to a certain degree, automatically can protect himself against recurrence of severe symptoms of the disease. There is a degree of immunity, or a premunition to malaria, which develops concomitantly with infection. This point should always be borne in mind when entering new areas and attempting to secure clinical evidence relative to the presence of the disease. In the great majority of instances, in even moderately severe endemic areas, clear-cut history of clinical malaria may not be forthcoming unless the investigation happens to coincide with that period of the cycle of malaria when active transmission is present. Because of this "immunity" in old residents, in endemic areas where a search for clinical history and subjective evidence are the only means of investigating the presence of malaria, an epidemic among the newcomers may well constitute the first clinical indication of the presence of disease.

Although an immunity or premunition against the disease can be produced, little of practical field value in prevention has been accomplished up to the present by vaccines or protective serums.

## 2. EMERGENCY PROCEDURES OF MALARIA CONTROL

#### a. GENERAL CONSIDERATIONS

Under normal circumstances, malaria control operations would not be attempted without previous investigative studies. Under normal circumstances, control methods would not be attempted by employment of methods comparable to a clinician's shotgun medical prescription. Emergency situations, however, demand procedures of attack altered greatly from those recommended for normal times. Frequently an emergency situation

will demand a type of guarantee that as many men as is humanly possible be kept in condition to perform their duties to the utmost of their capacity, irrespective of cost or possible duplication of control procedures. Under these circumstances, and where cost is not a factor, no method known to possess a fair degree of useful application should be overlooked.

In instituting measures for malaria control under emergency conditions, the routine procedures must be modified to meet these extraordinary conditions. Attempt will now be made to outline procedures of investigation and control as they should be employed when demanded by exceptional situations.

As, under emergency conditions, time will be the urgent consideration influencing attempts to prevent clinical cases, this schedule of procedure is planned to institute all indicated measures at the earliest time.

Control measures, under these conditions, fall naturally into several main categories:

Procedures of a general nature to be used.

Procedures to be used of an emergency nature.

Procedures to be used if considerable delay in the area is contemplated.

#### b. PROCEDURES OF A GENERAL NATURE

Malaria discipline must have been instilled into all before arrival at the point where protection against the disease is indicated. Frequently this can be accomplished while travelling on ship. The essence of the state of being or attitude known as malaria discipline is born of complete education of all personnel in the personally applied measures of control and an understanding of the epidemiology of the disease and procedures used for its control and also a knowledge of maintenance of existing control installations.

Malaria discipline is the individual attitude of decision to control the disease by adequate use of appropriate procedures of investigation and control.

Pre-requisites of any individual or organization entering an uncontrolled area where malaria is present must be alertness and decision to prevent the disease, and a supply of indicated equipment.

Under emergency circumstances it is assumed that the prime aim will be to prevent as much malaria as possible with the greatest possible efficiency, even though some measures may later prove to have been un-

necessary, or reduplication of effort may later be evident, or some procedures may be proven not to have been based on a complete scientific pre-study of the situation. Under these circumstances, it is assumed further that all other considerations will be sacrificed to the immediate attempt to prevent the clinical manifestations of malaria within the group or contingent involved.

When planning to enter potential malaria districts, or when personnel are to be allowed on shore in such a locality during periods of transportation by boat, or are to be exposed otherwise to infection, they should be prepared sufficiently in advance by doses of suppressive medication to build the plasma concentration of the drug to a level adequate to suppress the disease. Once having entered such an area, suppressive medication should be continued thereafter as long as the active services of these individuals will be demanded.

When ships are used for transportation of personnel, the first step on arrival at the malarious district is to insure that the ship be kept free of malaria-carrying mosquitoes. If the ship can be anchored sufficiently far from shore to prevent the access of anopheline mosquitoes, not less than a mile and preferably more, the work will be simplified. If the ship is anchored close to shore or at a wharf, and personnel remains on board or visits the ship, spraying with pyrethrum extract should be performed twice daily throughout the whole interior of the ship. If personnel is being transported by any means of conveyance (trucks, trains, airplanes), and stop at infected points, the carrier should be sprayed, after departure, with pyrethrum extract.

Under the circumstances existing in war time, and with the moving of men from one locality to another, there may be frequently offered, from a chronological standpoint, periods of time, during transportation or at other times, when logically certain accessory procedures of malaria control and education can well be performed. These procedures will anti-date, in large measure, actual application of the personally applied procedures which must be utilized when whole organizations are exposed to active transmission of the disease. In accordance with the above concept, opportunity may be afforded, during period of travel of an assigned personnel before arrival at any point of destination, either proceeding to or returning from malarious districts, to treat adequately with an accepted and recommended anti-malaria drug all persons found with positive blood smears. Blood smear examinations should be made of all cases of acute fever, and cases proven positive for malaria should be treated. All persons proceeding from areas where malaria is known to be endemic or where an epidemic has recently occurred should be treated as a group, as thoroughly as possible, or maintained rigidly under suppressive treatment, if they are proceeding for active service in new areas or a district where vectors are present. This practice will tend to prevent relapses of malaria caused by strain, overwork, change of climate, altitude or temperature, change of habits of life; will assist in preventing new reservoirs of infection being

introduced into new areas, and, will, in all probability, maintain the majority of the personnel as an effective force.

### c. PROCEDURES OF AN EMERGENCY NATURE

Emergency procedures of malaria control deal largely with personally applied measures in contra-distinction to what is generally known as environmental control. Personally applied procedures include employment of aerosols, repellents, suppressive medication, protective clothing, mosquito nets, head nets, and use of common sense in refraining from introducing oneself needlessly into localities at times when predisposition to infection would be very high. Environmental control involves the use of general procedures, such as drainage, fill, and formal mosquito proofing of buildings. Larvicidal application also falls into the category of environmental control; although, at times, it will have a specific use under emergency conditions. Drainage is also an environmental control, with no application to personally applied measures, but it, too, will have its uses under emergency conditions in active theaters of operation and combat.

All equipment necessary for the entire unit should preferably be transported with each unit of personnel in order that the units or individuals given special assignments may be equipped at time of arrival at malarious destination. At times, all recommended equipment may not be available. Under these circumstances, use that at hand to the best advantage. In malaria control, it is necessary to improvise.

All personnel should be instructed, prior to arrival at their destination regarding the manner in which malaria is contracted.

The common means of personally applied prevention and control should be outlined, and all persons instructed to use these measures. Supervision should be given to assure their use. Such procedures would include:

Keeping as much of the body as possible covered with clothes.

Hand-killing of mosquitoes inside of habitations, rest buildings, etc., both morning and night, and anytime during the day when they are seen.

Use of aerosols in these buildings on routine schedule.

Before occupancy of buildings, spray-killing of all habitations and buildings in malarious districts previously used by the resident population or other contingents.

Avoidance after dusk and until daylight of unscreened buildings, in malarious districts where people resident in the community congregate, such as moving picture houses, restaurants, saloons. (Note:

Arrangements of daily leaves, insofar as possible, so that this period corresponds to daylight hours).

Remaining inside of screened quarters from dusk until dawn.

Use of mosquito bars, if available, when sleeping.

Taking of the suppressive medication recommended in adequate dosage. (This should be taken at a regular time each day).

Use of repellents sprayed every three or four hours if possible on clothing and exposed portions of the body of persons assigned to special duties where there exists special predisposition to bites of mosquitoes, e.g., soldiers assigned to patrol or guard duty.

Use of repellents on the hands, arms, face, and neck, under any circumstances, when exposed to the bites of mosquitoes, and sleeping preferably with clothes on when sleeping in such localities.

Use of mechanical devices for protection against mosquito bites, such as head nets, mosquito boots, gloves, etc., when not contra-indicated by the nature of the assignment.

Selection of sites for permanent camps, and especially temporary sites, in localities as far removed as possible from other habitations of the resident populace. (Anophelines may be present there, but they probably will not be infected with malaria).

If the unit is to be an ambulatory one, moving rapidly from place to place, little more can be recommended than the procedures outlined for all personnel before arrival at destination and the equipping of each individual with the preventive materials and supplies.

The following minimal equipment is recommended as advisable for all personnel when travelling alone, or suitable quantities for units of personnel when travelling or stationed together:

Pyrethrum solution and spray gun, or pyrethrum-freon-bomb, (if available), in sufficient number for length of assignment in malaria district.

Mosquito nets for bunks, if living quarters are unscreened.

Sufficient anti-malarial suppressive medication should be provided to cover the period of assignment if personnel is stationed away from source of supply. Repellent solutions or greases will assist in protection. They, of course, should not be relied upon alone, but in conjunction with other measures will prove beneficial.

If the unit is to be exposed in populated areas, resident personnel

should be employed in and around the bivouac area with caution. These, though showing perhaps no clinical signs of the disease, may be parasite carriers and may serve as a source of infection to anopheline vectors around the base which up to that time have not acquired parasites from an infected person, and which could transmit a newly acquired infection in two weeks time.

Blood smears should be taken from such resident personnel before they are allowed access to the camp, and, if positive, they should be immediately isolated and treatment instituted. Blood examinations should be repeated on these individuals, preferably at weekly intervals, as a routine procedure, and also at the first sign of any sickness. If any of these people become sick with fever, or if parasites are found in the blood smears, they should be isolated by the use of a mosquito netting at least and, if parasite positive, treatment given.

If shelters or houses of the locality are to be used by the new arrivals, these should all be sprayed with pyrethrum extract before such use in order to kill any infected anopheline vectors that may be present. Especially does this procedure apply to places of congregation, such as moving picture houses, bars, restaurants, etc.

If shelters or houses are to be used for special purposes, where personnel will be particularly exposed to the bites of mosquitoes, such, for example, in military circles as shelters for patrols or guards, sprays should be provided for killing adult mosquitoes within the shelters, and repellents supplied for spraying the clothing worn and for application on exposed skin surfaces. Head guards with nets, if not contra-indicated by the nature of the assignment, may also prove useful.

When tents are used, formal mosquito proofing then being more difficult, reliance must be placed largely on sprays employed at least before retiring, and preferably twice daily, once before using the tent for retiring, and once later during the night. (At times, when pyrethrum is not available, a mess boy may be delegated to the task of hand killing all mosquitoes inside of tents or buildings).

All personnel should be instructed not to visit in resident habitations or buildings between the hours of dusk and dawn, and as seldom as possible at any hour. When such visits are unavoidable, personnel should be instructed to remain inside as short a time as possible.

The personally applied measures above noted should be employed rigidly.

#### d. PROCEDURES WHEN CONSIDERABLE DELAY IN THE AREA IS CONTEMPLATED

For actual works of investigation and control, it is recommended that each unit, destined for such investigation, and supervisory activities, should consist of, at least, a malariologist, preferably a physician, and

entomologist, and an appropriate number of skilled and unskilled laborers. If drainage measures are to be considered, or special activities of water control contemplated, an engineer must be included. The size of the area to be covered, considered together with the time allowable for such coverage, will determine the number of personnel in each classification of the unit. These determinations should be reached, and corresponding arrangements made by the responsible office before the unit starts for the point of final assignment.

If the assignment is of sufficient importance, and malaria is present, the physician (malariologist) should be assigned to the sole task of its prevention. His entomologists should be sufficient in numbers to supervise the preliminary works of control, and to cover the essential entomological investigations of the area in its main features, and institute preliminary environmental control measures within a period of not more than two weeks after arrival.

The first step immediately after arrival at the area of assignment will be the selection of the location for a camp and habitations, "habitations" having special reference to the point at which the personnel is to sleep or spend the night. In all probability this selection will have to be made immediately and accordingly cannot depend on an anopheline survey to indicate places where anophelines do not exist, or where breeding localities, which are within flight range, could be easily controlled. Where time does not permit of such entomological studies, the camp should be located in an area practical for military purpose, and at the same time, most distant, a mile or more if possible, from all human habitations of the resident population. Prevailing winds should be considered, and camps located on windward side of any recognized potential or actual breeding areas. The malariologist or entomologist, together with the malaria engineer, should select an area with a mile radius around it on high ground, if existent, as free as possible from potential breeding areas, and where water areas of the entire terrain involved (namely, camp site and the mile radius around it) is easily drainable to spots of lower elevation, where anopheline breeding would constitute no menace.

If possible, all habitations, offices, meeting rooms, and similar points of assembly should be mosquito proofed as well as possible with available materials, remembering that screen doors should open outward.

Latrines, a predilect spot for acquiring infection because of exposure of the body to bites and because of their use at night, should be as completely mosquito proofed as possible and provided with pyrethrum sprayers.

If the station or camp is to be semi-permanent or permanent, the entomologist should begin immediately the entomological inspection of the terrain comprising the station itself and a one-mile radius surrounding it. The investigations should be started at the location of the building themselves, and as anopheline breeding areas are located, larviciding crews should be sent with the entomologists who should supervise the application

of the larvicide. In the meanwhile, the entomologists should continue the search for new breeding areas, radiating out from the habitated area to the perimeter of the mile zone.

The attack against breeding of the vector is designed to relieve the continued necessity for constant application of the personally applied measures of control, which, with long use, become onerous.

No special study should be directed, during the control measures of these first days, toward establishing the identity of the vector or exploring the possibility of species sanitation. Such scientific approach, entailing special study, must be postponed until all anopheline breeding areas identified by the entomologists are treated. This should be accomplished as quickly as possible. The temporary omission of detailed investigative procedures is recommended in order that the real vectors of the region may be controlled immediately, although, possibly many species, later proven to be innocuous, may at first be included.

As time permits, and if the station is to be used for longer periods, maps should be prepared, and all breeding areas located on them. This procedure facilitates the routine weekly larviciding of such areas.

Once principal breeding areas have been located, and larviciding or minor drainage, if practical, instituted, the entomological service should include in its schedule of activities the routine collection and identification of the anopheline fauna of the locality, and the localization of the areas, and the determination of types of areas in which different species breed.

Qualitative and quantitative information regarding the mosquito fauna is secured by means of mosquito capture stations, mosquito traps, and larval collections. From the facts revealed by these captures, relative to the successive adult captures and their densities, effectiveness of larvicidal operations may be measured. Evidence gained through larval collections, performed as a "pre" and "post" larvicide application will also assist in the evaluation of control measures employed. Catching stations should be strategically located, especially in the neighborhood of the living quarters, as well as in relation to individual breeding areas, and as a meshwork covering the whole locality.

When the program has progressed and time permits, anophelines should be captured inside or near houses. All of these, especially blooded anophelines captured in the vicinity of habitations, should be subjected to the procedure of dissection, observing the precaution of maintaining the blooded anophelines alive for as long as eight to ten days if possible before dissection; so, affording time for any recently acquired infection to develop within the mosquito. Mosquitoes, so kept for dissection, should be inspected at frequent intervals, and any found dead immediately dissected. In the majority of instances, however, the vectors of areas are already known and mosquito dissection would be performed for academic or research

purposes.

If and when actual cases of malaria are discovered among the residents of the locality, or the personnel of the group, their habitations should be visited for the capture of adults for dissection. Anophelines captured under these circumstances would comprise the group of mosquitoes most likely to harbor the organism. Through practice of this procedure, the actual vectors, when not already known, would be more easily identified. Also, under these circumstances the houses, after collection of mosquitoes is apparently complete, should be sprayed with pyrethrum.

Through dissection of adult anophelines, and demonstration of infection in salivary glands and stomachs, it is possible to identify the species, if not already known, which are responsible for transmission and, through the correlated entomological studies which reveal actual breeding spots of different species, species sanitation may be instituted. Application of species sanitation can result in considerable saving of time and money, and prevent undue exposure of military personnel engaged in investigation and control, since areas, not breeding the actual vector, can be eliminated from areas to be controlled.

If and when time and personnel permit, blood smear diagnosis should be practiced on native persons residing near or having constant contact with the personnel of the recently-arrived contingent, such as laborers at the camp, and all cases having parasites should be adequately treated in order that reservoirs of parasites may be eliminated. The houses where these people live, which they frequent, or in which they congregate should also be sprayed with aerosols. In association with the study of the parasite it should be remembered that the gametocyte stage of the parasite, which is responsible for the infection of anophelines, is at times the stage of the parasite most difficult to influence by medication.

If the station is to be semi-permanent, it is recommended that the malaria unit should have available, as minimal reference books, certain publications for the use of the whole unit, namely:

- (1) Human Malaria, American Association for the Advancement of Science, Smithsonian Institute Building, Washington, D. C., 1941.
- (2) Key to the Anophelines of the World, Russell, Rozeboom, and Stone, Published by the American Entomological Society.
- (3) An Introduction to Malariology, Dr. Mark F. Boyd, Harvard University Press, Cambridge, 1930.
- (4) Manual for the Microscopic Diagnosis of Malaria in Man, National Institute of Health, Bulletin No. 180.
- (5) The Anopheline Mosquitoes of the Northern Half of the Western

Hemisphere and the Philippine Islands, published at the Medical Field Service, Carlisle Barracks, Pennsylvania.

- (6) The Mosquitoes of the Southeastern United States, King, Bradley, and McNeel, U. S. Department of Agriculture, Miscellaneous Bulletin No. 336, Revised June, 1942.

### 3. SPECIAL CLINICAL ASPECTS

When any degree of permanency in the area is anticipated, unless the area be known to be either highly malarious or actually non-malarious, the malariologist should investigate the actual status of malaria among the residents of the community. However, as noted above, some practical activities must precede this investigation. As previously noted, a site for a camp must, of course, be selected immediately upon arrival if a base, even temporary, is to be established. This must be done to the best of one's ability, and when the presence of malaria is known or suspected, in accordance with recommendations cited above. Also, as an insurance and a safeguard, the entomological program for larval control should be instituted when malaria is even suspected.

In many localities where malaria units will be operating, history of the disease, and mortality and morbidity statistics will probably be non-existent, or of little use and accordingly the important investigations to determine the presence, intensity and phase of the disease will have to be objective in nature. The splenic examinations with associated blood findings will offer the best approach to the clarification of this subject.

Findings of the splenic examinations are immediately available, and will define the general status of malaria in the community, and its geographic location. Of course, in tropical countries, care must be exerted to rule out other causes for splenomegaly. Blood smear examinations, when positive, will be absolute. These necessitate, however, a relatively long period for their examination.

The spleen becomes enlarged in certain acute and chronic diseases. Acute diseases are characterized by a rapid decrease in splenic size to normal when the disease is cured. Splenic examinations should be avoided during the course of epidemics of acute febrile disease, other than malaria, and for a period of approximately a month following the cessation of such an epidemic.

Finding of a considerable percentage of enlarged spleens in the presence of no acute disease, or recent history of acute disease, is usually significant of endemic malaria.

The presence of splenomegaly, attributable to malaria, will establish the potential danger of transmission of malaria to the newly arrived personnel. It should be remembered that the enlarged spleen is evidence of a developed, or developing immunity to the disease; whereas, parasites in the

peripheral blood indicate recent transmission, or cases which have recently been active.

Enlarged spleens substantiated by the discovery of malaria parasites in a considerable number of these same cases naturally confirm the general splenomegaly as surely indicative of malaria. When enlarged spleens in considerable percentages are found and not confirmed by parasites, the assumption is still that the splenomegaly is caused by malaria, unless other causes for splenomegaly in the area be known, or be demonstrated. Parasites are not always present in sufficient density to be demonstrable, at all times, in the peripheral blood of cases harboring a latent malaria infection. In certain tropical regions, kala azar as well as schistosomiasis exist, and can cause a splenomegaly which can be confusing in some localities. Otherwise, there should be little cause for confusion when large numbers of cases are found with splenomegaly in the absence of the actual presence of recent history of acute disease. (Note: Other causes of splenomegaly are hereditary syphilis and rickets. Of course, various other diseases can cause splenomegaly, but the likelihood of their being present to a degree which would cause any confusion is remote).

The malariologist should a priori consider all cases of acute or continued fever occurring in a malaria district to be possible cases of malaria and should take blood smears daily for diagnosis until the case is established as positive or, through repeated negative blood smears, the conclusion is justified that the case is negative. Such febrile cases should be isolated, if possible, in a screened room, or at least, by use of a mosquito net. It should be remembered that in first days of fever, in cases with few previous infections with malaria and little resulting immunity or pre-munition, sometimes parasites are not present in the peripheral blood in sufficient quantities to be easily demonstrable. When the disease is further advanced, such diagnosis is easier. When cases of undiagnosed fever occur among those taking suppressive medications, if the individual is not essential at the moment, these medications should be withheld for a period of days in the hopes of establishing a definite diagnosis by means of blood smear examinations.

The malariologist should always remember that malaria can be introduced through humans, reservoirs of the parasites, into an area which hitherto has not suffered from the disease. Vectors may be present in the locality, but previously there may have been no reservoirs of parasites. Furthermore, an area which has been mildly endemic, because of a developed immunity among the residents to a particular strain of parasite, may be made highly epidemic by the introduction of a new strain of parasite; or, epidemics may result in such a community among the recent arrivals who may be non-immune to that particular parasite which is already existent in the community, and to which the populace has become largely immune. For these reasons, it is recommended that persons coming from one malarious area, and proceeding to another where vectors are present should be at least kept under the control of suppressive drugs if not treated thoroughly.

Military personnel travelling quickly from one district to another constitute an extreme hazard for introduction of new parasites to a new area. In this connection it may be useful to recall that the incubation period of the parasite in the mosquito varies under different circumstances, especially, with varying temperatures, but the average may be stated to be approximately ten to fourteen days.\*\*

The incubation period of the disease in man is also variable, depending on certain circumstances, including the species of infecting parasite, but may be stated generally to be also ten to fourteen days.

Gradually, and if the station is to be continued for sufficient time, the scientific approach to the study of the disease should be established. This program offers a scientific background for control, and makes the program more economical, and more thorough. Such studies, postponed temporarily for urgent reasons, when instituted, may result in a control limited to those areas, only, responsible for the production of the active vector of the disease.

\* When, in a permanent or semi-permanent base, the work of control has progressed to a stage where larval collection and adequate adult captures indicate conclusively that vectors are not present at any point where the personnel is stationed or where persons may visit, suppressive medication may be suspended until personnel be given assignment in an area where control is not efficient.

\*\* At temperatures ranging from 25° to 30° C., the incubation period in mosquitoes is eight to nine days; whereas, at temperatures between 15° and 22° C., it is lengthened to eighteen to nineteen days.

## ENTOMOLOGICAL PROCEDURES IN RELATION TO MALARIA INVESTIGATIONS

For an understanding direction of application of malaria control procedures, it is essential to have a clear knowledge of the basic procedures of a malaria survey, ability to compile the data secured, to analyze it, and, finally, a realization of the significance of the analyzed data. At times, circumstances may be such that it will be impossible to perform a complete survey. However, under such circumstances an understanding of the procedures employed, their use and significance, will enable one to select those special investigations which are indispensable for an effective application of available control measures.

A few examples will suffice to illustrate the validity of this contention. In hyper-endemic areas the dose of suppressive medication would be necessarily larger than that actually sufficient for effective suppression in areas of low endemicity.\* In areas where P. falciparum is the common infecting parasite, high mortality rates may ensue unless the nature of the infecting parasite is known and its import realized. The type of control against the vector and the urgency of it will depend on a knowledge of whether the vector is sylvan in its habits or highly domestic or a combination of both. Furthermore, it is known that certain species of anophelines are avid vectors of the parasite, such, for example, as A. gambiae and A. darlingi. In order to eliminate the "seed bed of infection", gametocyte carriers and parasite carriers must be recognized and their place of residence known. Such points as these are revealed only by the employment of the appropriate isolated investigations included within the general framework of a complete malaria survey.

It is true, as previously noted, that personally applied methods of control and mosquito proofing do not necessitate, for their justifiable application, further knowledge than the fact that malaria is occurring in the locality or perhaps that it can occur. However, the first approach to relinquishing alertness in the application of such tiring procedures of personal application, and substitution of more permanent measures, demands utilization of investigative methods included in surveys.

Under any system of control procedures being employed, the misdiagnosis of a hyper-endemic area or ignorance of the common presence of P. falciparum might cause serious disaster.

From the side of clinical investigation, facts should be known other than the presence of the disease, its location, its spread, and intensity, and the phase which is existent in the locality at the moment.

\*Note: 0.1 gram of atabrine per day, 6 days per week, when actually taken, will suppress clinical symptoms of P. vivax and P. falciparum malaria under hyper-endemic conditions and with P. falciparum the suppressive dosage, if continued for a month after new infection has ceased, will frequently cure the disease.

The parasite and its life inside the human host, with its proclivity for relapses, must be understood. The capability of the human host to defend, by his own protective reactions, the body against parasitic invasion must be appreciated, as well as the importance of the sexual cycle in man in its potential role for infecting mosquitoes. Eliminate the parasite, or better stated, the sexual phase from the human reservoir, and malaria would cease.

The cycle of the life of the parasite inside the body of the mosquito is of equal or greater importance from the standpoint of control since the life of a mosquito is limited, and accordingly, the life of the parasite inside the mosquito as well. The factors which favor or curtail the development of the parasite inside the mosquito, and the factors which promote or militate against the life of the mosquito itself are of the utmost importance in the cycle of the propagation of the disease. Sufficient rainfall, warm climate, humidity, in correct proportions and correct degrees, are favorable to the life of both the mosquito and correspondingly the life of the parasite; whereas, opposite conditions present an unfavorable environment. Hyper-endemic situations, for instance, cannot exist where meteorological conditions do not favor the production of the vector in sufficient densities during a considerable minimal portion of the year. All phases of the inter-related life of the mosquito and the parasite, as well as the life history of the anopheline vector itself, should be understood for formulation of intelligent plans for the control of the disease. It must be remembered that the propagation of malaria is an accident occurring in the life history of certain anopheline mosquitoes, and due to the habits of the anopheline vector.

Entomological studies are complementary to and a natural correlary of the clinical studies of a malaria survey. That malaria is existent is revealed by the clinical survey. Further, the survey reveals, to a partial extent, the reason for the dissemination of the disease in that locality, as parasite and gametocyte reservoirs have been discovered and the place of abode of persons harboring them can be noted. At this point, entomological investigations, revealing breeding areas and their location and relationship to the seed bed of parasite, as well as the life habits of the vector, must complete the picture.

The actual vectors, their breeding places, flight range, bionomics, their susceptibility to infection, their pre-dilection for human blood meals, their habits of domesticity, their preferred time for biting, are among some of the points that must be clarified by entomological studies. Frequently, by mere positive identification of a vector, these facts will be known, as the species may have been thoroughly studied, and all facts relative to its capacity for transmitting the disease adequately recorded. At other times, special and perhaps original studies must be made to verify these points.

Entomological studies afford a general orientation on the anopheline fauna of the region, and are designed to elucidate, as well the subject of

densities and seasonal variations. When it is possible to continue such investigations for sufficient time, they may perhaps show cyclical variations in breeding over a period of years.

The investigation of the ecology of species may reveal points of attack logical for their control. Preference for various breeding conditions once known may be altered, and by such artificial changes, breeding may be naturally curtailed if not eliminated.

In addition to yielding general information on the fauna of mosquitoes of the region, and affording a general orientation, entomological studies, both on larvae and adults, serve as an excellent yardstick to measure the progress of control works. Increase in densities of the vector or vectors in a malarious district will serve, before the clinical advent of the disease, as a danger signal of heightened entomological activity, and the possibility, even probability, of increased transmission of malaria. Larval inspections indicate whether there is need or not for application of larvicides when inspections are made prior to and after larvicides have been applied.

A capture station is any spot or collection of spots chosen for the capture of mosquitoes. With reference to malaria, the mosquitoes being investigated are naturally anophelines. The station may consist of such localities or constructions as latrine, stable, house, underneath a house, culverts, or may consist of a combination of these all in the same limited area.

A mosquito trap is any device used to attract mosquitoes, and designed to prevent their easy exit. The classical traps utilized in the investigation of mosquito fauna are: Animal bait traps, humid box traps, blackened box traps, light traps, human bait traps, mosquito net traps with two nets.

Whatever system or combination of systems are used, employment of capture stations and traps constitutes an attempt to sample the mosquito fauna, and by routine collections consecutively performed at suitable intervals, secure information regarding density, at least relative, if not absolute.

These studies are accomplished by the use of adult capture stations, and mosquito traps, both located at diurnal resting stations in the vicinity of known breeding areas, as well as at spots frequented by anophelines in the search of a blood meal. Larval investigations and collections also constitute a part of such studies. Comprehensively, the studies involve investigations of the important factors associated with typical breeding areas, field observation to determine the period necessary for development from egg to adult, stained specimens, or comparable procedures, for ascertaining flight range, precipitin tests for demonstration of blood meal preference, and accordingly, the source of blood meal associated with the breeding area, and dissection for demonstration of vector of the area.

With a good observer, larval inspection can be sufficiently objective to be relatively accurate. Unfortunately, adult captures, accomplished by capture stations or by means of traps, are much more subject to variations and vagaries. The reason for this is obvious, and hardly needs comment. Larvae are, by their nature, confined to water areas, and the capacity for self locomotion is not highly developed; so, once in a district, they remain in the same general area until the stage of adult is reached unless moved by outside agencies. Capture of adults is only as successful as the ability of the entomologist to locate a place attractive to the adult where such favorable factors continue to persist. As a general rule, such a place would be one characterized by shade, dampness, protection from the wind, and probably, associated with the proximity of the preferred blood meal. They should, in general, be located somewhere between the source of breeding and the closest source of a suitable blood meal, and, of course, in the area to be protected, here serving as a measure of the effectiveness of control measures. Positive findings, at a capture station, are definite though not necessarily all inclusive, either for the species present or their absolute densities. Negative findings, at station or in traps, may not present evidence that mosquitoes do not exist, but may indicate that the station has not been well located, or that factors essential to the attraction of mosquitoes to that locality may have become altered since the time it was originally established. Subsequent to the original selection of a capture station, trees may have been felled and protective underbrush cleared; prevailing winds may have changed, resulting in exposure to these, rather than protection from them; humidity may have been altered by planned drainage or natural drying; a previously shaded protected area may have been converted into a sunlit plain. An area may be converted from a secluded spot, perhaps, into a junction of railroad lines, or an industry, repellent to mosquitoes may have been established in the close proximity to the station. A ready source of a blood meal, a dairy for example, may have been removed. Negative captures under conditions altered since the station was established, and where its effectiveness had been previously proven by positive findings, would not necessarily indicate a reduction in mosquito presence, nor indicate that such apparent reduction was attributable to application of control measures. Under these conditions the negative results would simply demonstrate that an adequate and representative sample of the existent population was no longer being secured by that station. For these reasons, it is obvious that all fundamental factors, favorable for the presence of adult mosquitoes at capture stations, or traps, should be recorded when the station is established, and any change therefrom, noted if the station records a decline in captures or produces only negative results.

It is proverbial that anophelines generally bite at dusk or predawn or during selected periods of the night. The entomologist, accordingly, should be on the alert for studies conducted at night, and not place all reliance on daytime findings. Many essential habits of these mosquitoes may be revealed only by night time studies. Perhaps the most important practical point in the life habits of anopheline mosquitoes is related to

those habits which are intimately associated with the acquisition of, and transmission of the malaria parasite. The time of flight, densities, permanence inside of houses, resting after biting inside of habitations, or immediate exit after biting, abhorance of habitations, preference of choice between animals and man for blood meals, the number of times the anopheline will take a blood meal before the extrinsic incubation period has matured; all of these facts may only be comprehensively revealed by special studies conducted at night when the vectors are actually at the work of seeking their blood meal, and, incidentally, from their standpoint, transmitting the disease to humans.

Whatever shortcomings may exist intrinsically in securing adequate sampling of the anopheline fauna through adult captures and larval collections, at least, procedures can be planned and adopted whereby the results should be quantitatively comparable. After preliminary trials to ascertain if stations actually secure a sample of the locality, a routine of observations should be established whereby such samples may be considered to be comparable. Ordinarily, when time and personnel permit, captures are made at weekly intervals, and preferably at the same time of day and night with the same observer making the observations in his particular district. Meteorological findings at the time of collections or captures should be recorded.

Whatever system be adopted for measurement, whether in the case of larvae, per square meter or average of dips, or any other estimation, the same system should be routinely pursued so that results of density may be comparable. The same may be said regarding adults. These records may be standardized in accordance with total capture of the same square foot surface, or per square meter, or by man hour, or by killing with an aerosol and total capture, or by estimation, or by total capture made by suction or chloroform tube. Here again, whatever system be decided upon, and finally adopted, this system should be continued thereafter without alteration. Insofar as may be possible, the same observer should continue the routine captures and larval collections at the same stations. By this procedure, the results will be more comparable. At times, check up inspections of areas by different observers may be indicated, but this would not influence or change the routine procedure of observation made in the area.

## ENGINEERING PROCEDURES IN RELATION TO MALARIA INVESTIGATION AND CONTROL

Engineering procedures in malaria investigation and control comprise the study and execution of certain specific means of control which involve largely the disposition and treatment of water areas breeding the anopheline vector. Although others than engineers may have information of a general nature sufficient to enable them to treat of elimination of simple problems relative to anopheline breeding areas, the engineers should be entrusted with the study and execution of any program of elimination involving difficult situations.

Application of engineering procedures to control of water areas is necessarily closely linked to, and dependent upon, entomological findings. The expenditure of labor, time, and material, for instance, to control a breeding area two miles away from the area to be protected is not justifiable if the effective flight range of the anopheline vector in question is limited to one mile.

Removal of all exposed water areas where anophelines breed, would eliminate malaria. (It is, however, readily apparent, both from an economical and physical standpoint, that except in definite limited areas, the removal of all breeding waters is impossible. Therefore, alternate measures must be taken to control exposed water areas, and render them unfit for breeding purposes).

Among the procedures, accessory to drainage or fill of water areas, which the engineer will be called upon to study and perform, as these relate to potential or actual breeding of the malaria vector, are the following: Management of impounded water areas, chemical or organic change in character of waters, use of tide-gates, flushing, agitation of water, retention reservoirs, installation and operation of manually operated gates, diking, surface grading, and finally, generically speaking, the most important procedure of all, the general consideration of the ultimate and effective disposition of residual waters.

According to the belief of some, larviciding and the use of predators should also be a function of the engineer. However, in view of the fact that the criterion of success of such procedures is based upon and measured by constantly repeated larval observation, it is believed by many that these procedures would fall more logically within the province of the entomologist.

Any water surface, natural or man-made, may be a mosquito breeding area. Natural water surfaces include rivers, streams, lakes, ponds, swamps, marshes, springs, and shallow depressions in the ground surface which collect and hold surface runoff. Man-made water surfaces include reservoirs and other impounded areas, wells, cisterns, borrow pits, ruts, bar-

rels, tin cans, etc. Many of these man-made breeding areas are the result of carelessness and lack of foresight. Accordingly, they are avoidable. Though sometimes the point is overlooked, prevention of the formation of water surfaces is in itself a control measure, and one that pays dividends in the savings of labor, time, and sometimes materials. Engineers themselves, in many instances, are the most guilty of infraction of this concept. Borrow pits are landmarks of most construction works. In any location they may collect and hold water, and in low-lying areas are impossible to drain and expensive to fill. Borrow pits should be located on ground that is sufficiently high so that subsequent drainage is possible, and, wherever possible, the earth should be "borrowed" in accordance with a survey which results in surface drainage. Highway and railroad engineers, thinking of flood water removal or equilization of hydrostatic pressure, are oftentimes guilty of placing culverts at elevations adequately placed for the care of flood waters, but too high to afford drainage of residual waters, resulting in impoundment of residual runoff at the site of the culvert. Those two examples are classical. There are many others.

From the standpoint of malaria engineering, storm water and surface run-off from a water shed is of interest, only insofar as such waters may cause residual waters capable of breeding the anopheline vector. It is natural that engineers without specific training in malaria control procedures should accentuate in their work the removal of these storm waters to the practical exclusion of consideration of residual waters since waters, resulting from flash flooding, have always constituted a main problem. The engineer, trained in malaria control, soon learns the importance of other bodies of water which exist as residual caches capable of breeding anophelines.

The procedures of drainage utilized in malaria control operations can be classified in various manners from the standpoint of the type of water which is removed, as well as from the viewpoint of the type of drain designed to remove it.

In accordance with the first classification, water can be divided into storm water, surface runoff, residual water, and sub-surface water.

When drainage is classified in accordance with the type of system removing the water, it can be sub-divided as follows: Central drains, lateral drains and intercepting drains, and these classified again into open drains and closed (or underground drains); lined drains and unlined (or earth drains). Vertical drains are in a category by themselves, and will be discussed briefly later.

Generally speaking, open drains are cheaper and easier of construction than closed drains, but the maintenance on them is obviously much greater. They are constantly subject to the possibility of deposition of various materials and objects in the drainage line, and the resulting impairment of their intended function. Open drains also, by their nature, constitute a handicap to the ordinary use of the areas where installed as they can

only be crossed with ease at points where bridges are established. As a general rule, open drains will necessitate a higher gradient than closed drains to drain the areas efficiently. The reason for this is obvious. Minor obstructions are constantly being introduced into open systems, and then can constitute a blockage of the drainage unless there be sufficient head of water to create a velocity of water flow, which is capable of washing out and cleaning the systems. The closed system is not as readily subject to such casual blockage since foreign objects are not as easily introduced and sand traps and observation boxes are installed in the system for the purpose of arresting any such casual objects or material, as well as to assist in the work of ordinary maintenance.

An unlined ditch will remove water and can eliminate breeding areas. Its importance as a temporary expedient should never be under-estimated. Here again the gradient of the unlined ditch must be greater for efficient and comparable function than that of a lined ditch. Maintenance in the unlined ditch will be excessive as compared with the lined ditch. The design of such an open, unlined channel should be made with due consideration of the type of soil through which it will function. Soft loose soil will wash out readily so the grade should be adapted to produce a sufficiently low velocity flow to prevent scouring or erosion, and the side slopes kept well within the limits of the "angle of repose". As soil types change from sand to loam, to clay, increased grades are allowable with resulting increase velocities, and the "angle of repose" can approach more the vertical. Limiting grades for unlined ditches (except through rock) are generally conceded to conform to gradients of 0.05% - 1.5%. In closed concrete lined systems gradients as low as 0.01% (1 foot per 1000 feet) can be used in a carefully installed ditch line. Gradients chosen should be based, not only on the type of soil through which the ditch passes, but also should be adapted to the knowledge of the lowest elevation of areas necessary to be drained in the upper portions of the system and with due consideration for resulting cuts which the proposed gradient will necessitate. Generally speaking, a central ditch should be established at the lowest effective elevation. Under these circumstances, it will serve for all areas capable of being drained in the area where drainage is planned. If it becomes necessary to change gradient proceeding from the upper reaches of the drainage system to the lower, it is always safe to change from a lesser to a greater grade. It is not advisable to change from a steep gradient to a shallow one directly. If it is necessary to change from a higher to a lower grade as the drainage system proceeds down grade, it is always advisable to pass the water through a sand trap before discharging it into the line of lower gradient. This eliminates the possibility of sand and debris settling out in the drain line of lesser gradients because of the slower velocity of flow occasioned by the lower gradient. It should be remembered that a pipe on a shallow gradient has less carrying capacity than a similar pipe on a steeper gradient.

In areas where roots of various growths exist, it is not advisable to use subsoil tile line of less than six inches in diameter. Smaller sizes tend to clog too easily.

Tile lines should be laid out by someone competent to handle a surveyor's level. Grade must be exact.

Under conditions of emergency malaria control, possibly the open unlined ditch will be the one most frequently employed. The employment of open, unlined ditches does not absolve the malaria control unit from the responsibility of making these ditches as maintenance free as possible, and as effective as possible for the removal of water and with the ultimate idea in view of installation of linings if the ditches are to be continued in use as a semi-permanent or permanent installation.

Open ditches may be lined in various manners, probably the most satisfactory being the use of concrete products of either the pre-cast or monolithic variety. They can be rock lined or lined with rubble inverts.

With reference to the real merit of pre-cast and monolithic products, pre-cast products have certain merits as compared to the monolithic poured-in-place structure. They can be manufactured during periods of slack work or inclement weather. They are easy of installation and setting to proper grade. They can be installed on tressels through a boggy area. Repair and re-alignment is simple. Seepage lines, when the joints are left ungrouted, afford much more seepage possibilities for entrance of residual water into the system than is provided by the weep-holes of monolithic structure, which are incorporated mainly for relief of hydrostatic pressure. If, and when, it is determined to change a system from open to closed, or to remove a system entirely, pre-cast products can be easily removed, and utilized elsewhere if desired. They can be easily repaired by the use of new pre-cast sections. If it be desirable to utilize a system as an aqueduct for transporting water through an area rather than to lower the water table in that area, pre-cast units can be grouted together and a resulting monolithic structure formed. The drainage capabilities of pre-cast products left ungrouted in lowering water elevations is limited to the invert elevation of the bottom of the invert. In monolithic structure this limitation of drainage and lowering of water elevations is limited to the top of the side wall, with the exception of the small drainage facilities, which are afforded by the location of weep-holes placed at lower elevations within the structure of the system.

Closed or underground systems can be installed with concrete pipe ranging from four inches to several feet. The tongue and groove style of pipe in the larger diameter has proven very satisfactory in malaria drainage. The joints between the pipes are grouted or wiped on the top half with mortar, and the bottom part of the joint is left open (ungrouted). A good practice to follow is to place gravel around the joints of two pipes as the pipes are laid in place. This facilitates free entry of water around the lower half of the pipe, and prevents, to a large extent, the entering of silt through the bottom of the pipe, whereas, the grouted top prevents the entrance of silt at that point of possible entry.

Pipes are made of various materials. The type of material will be, to

some degree, dependent on the location and ingredients and equipment available. A good concrete pipe has proven to be very effective, made from sand, gravel, and cement, with water in appropriate mixes. The utilization of a dry mix should never be overlooked, as through the employment of this mix, one pipe mold can be re-used immediately after the product is tamped in place. This reduces the quantity of equipment necessary for producing pipes or inverts.

Also, the procedure of making thirds of a full round, which serve as invert linings, is possible through placing of pallets in an upright pipe form, and utilization of a dry mix. The pallets are held perpendicularly in place by holes made in the outer shell and inner core, and a corresponding hole in the pallet itself. When the pallet is in place, a nail is inserted through the corresponding holes. Under these circumstances, tamping proceeds as in the manufacture of a full round, whereas, the resulting product, when tilted from the bottom ring, will be three equal portions of a full circle. The molds for manufacture of these forms, together with a contracting core, can be made of wood or improvised in other manners. (See diagram, pages 203 and 204).

It should always be remembered that the resulting strength of a concrete product is largely dependent upon the amount of water or "curing" which it may receive during the period immediately following manufacture. If facilities are not available for emersion in a curing tank, the product should be watered by spray, from a hose, or by lawn sprinkler for varying periods, depending on the climate, humidity, and rainfall of the area. As a general rule, two weeks to a month of watering, eight to twelve hours a day, would be sufficient to produce a product of adequate strength.

The drainage of flood waters or surface runoff is the type of drainage with which the average engineer has usually been engaged. Usually, training in the importance of drainage of residual waters for malaria control has been relatively neglected. Residual water drainage is the type of drainage in which the malaria engineer should be primarily interested.

Waters may accumulate on the top of the ground because of an impervious ground layer beneath. Residual waters may also be present in localities as a surface expression of the ground water table itself in that area. Accordingly, it is evident that malaria drainage is concerned with surface runoff waters, only insofar as they may cause residual waters capable of breeding.

However, probably the most important phase of drainage for curtailment of anopheline breeding is that phase which treats of the lowering of the ground water table in those areas where water readily expresses itself as surface water capable of breeding. Such a situation occurs when there exists a high persistent level of the ground water table. Drainage of the surface of the ground to a central drainage system by the practice of surface grading can eliminate a large portion of flood waters. Lowering the persistent ground water table to prevent expression of surface water capable

of breeding, as well as removal of excessive amounts of water absorbed by the ground at times of rain, necessitates a planned system of drainage. The area over which a system will be able to reduce the elevation of sub-surface water will be dependent upon the permeability of the soil and the depth of the drainage system, and accordingly, this extent will vary under different conditions. The more permeable the soil, and the greater the practical depth of the drainage canal, the greater will be the total area over which a system functions in lowering the water table.

Vertical drains are mentioned more for the sake of completeness than because of their universal application. Their function depends upon piercing of a subterranean impermeable strata, which holds the surface water, and reaching a permeable strata with a free outfall. When the engineer has the good fortune of finding such conditions, areas can be drained by vertical drains. There are several points, however, which must be borne in mind. Penetration of an impermeable strata to a water layer where a head of pressure from the surrounding perimeter exists can produce, instead of removal of water, an artesian well. Furthermore, efforts in vertical drainage may be successful during the first period of use when a contained cavernous area has been tapped capable of receiving a limited amount of water, but which, when later filled with drainage water, will be incapable of further drainage. Polluted waters may also be introduced by vertical drainage into a system of water used for drinking purposes. Accordingly, the practical application of vertical drainage systems is decidedly limited. On the Coral Islands of the South Seas, vertical drainage has been most successful in a number of cases. Portions of these islands are composed of a very porous coral formation which is overlain with a layer of black impervious clay, which ranges in thickness from several inches to twenty or twenty-five feet. The impervious clay layer can be penetrated by the well, and water readily flows into the porous coral.

In considering the drainage of any area, the first concern is the location of an adequate outfall, or place to discharge the water, where it will no longer constitute a hazard from the standpoint of health or for any other reason. An outfall should be placed at the lowest possible point compatible with the indicated drainage of the area for which it is to serve. If possible, it should be sufficiently low to allow drainage of all points of the area involved, as well as to allow adequate gradient or fall for efficient function of the drains between the lowest point in the area to be drained and the point of discharge.

If avoidable, outfalls should not be selected by the unaided eye.

If the area is large, a map showing prominent topographic features, as well as all water areas, should be prepared. This will assist in determining the natural location of the drainage line as well as indicate special spots needing drainage.

During the process of investigating the terrain for an outfall, or during the period of mapping the area, the location of the drain line will

probably become clear. This drain line should be as straight as possible. If bends are necessary, two procedures are possible.

1. Make the bend with as long a radius as possible.
2. Make an abrupt change of line by means of modified junction box or through a sand trap. (Discussed later).

During the search for an outfall, the type of soils common to the area to be drained should be noted. Much of the success of the drainage system may depend upon the ability to judge the type of soil encountered, and its relative permeability.

Generally speaking, soils can be classified roughly into three types. These types are: (1) sand, (2) loam, and (3) clay. Sandy soils are usually considered most permeable and clay soils least permeable. But, there are all sorts of mixtures of the three types and, accordingly, there may be great deviations from the expected.

If time is available, during preliminary investigation for drainage, the study of the question of permeability may be productive of good results.

For such a study, inspection wells may be utilized. If more elaborate wells cannot be installed, simple holes in the ground will suffice, about eight inches in diameter, and dug, with a post hole digger, as deep or deeper than the proposed ditch. Such inspection wells can be situated in rows at regular intervals of about twenty-five to fifty feet apart and perpendicular to or across the ditch line. They are extended away from the ditch line as far as observations on subsoil water elevations would appear to be indicated. Succeeding parallel rows may be separated at intervals of fifty or one hundred feet. Such wells are very useful in determining the action of underground water elevations under varying time relationships to known rainfall as well as for estimating the effects on such elevations accomplished by installation of drainage lines. Once the central drain is in operation, the elevation of water in the inspection wells, when recorded and interpreted, reveals the general location for additional drains to produce effective results and the necessity for installation of such additional systems as interception drains, surface grading, and lateral drains, (open or subsoil).

These wells would not be employed in preliminary efforts of malaria control drainage. However, they should be remembered. They may be useful.

Long poles, especially bamboo, properly placed in the drain line parallel to the direction of water flow, produce a reasonably good drain, as does a properly constructed underground stone drain. However, earth covered pole and stone drains are usually short lived because of their tendency to silt and clog. Herbage packed drains, located usually in shallow ditches, properly packed with overlapping coconut palm leaves, afford fairly good drainage where the runoff is constant, but slow. This type of drain is used success-

fully in the Malay States, especially Singapore.

Particular attention should be given to several special features of a formally constructed drainage line which are frequently included in the line and are additions to the drain line proper. The first of these structures is the junction box -- where two or more lines of tile join together. It is desirable, but not always absolutely necessary to have elaborate junction boxes. Where elaborate, they are usually indicated because of sandy soil. In a firmer soil, such as loam, such boxes may be omitted and the two lines of tile brought directly together through a V-shaped tile or through a tile properly adapted by breaking. One point, however, should be emphasized which is common to either method of uniting the line; namely, there should be a slight difference in grade between entrance and exit tiles of the lines. In general, tiles of the entrance line should enter the main drainage system at least one-tenth of a foot above the grade line exit of the main line, and at such an angle that the flow of the main system will be interrupted as slightly as possible.

As stated above, sand traps are necessary for permanent lines in sandy soils. Under these circumstances, such traps are essential in order to collect foreign material, especially sand, which is washed into the line with resultant stoppage of drainage. This material must be removed, as indicated, from the sand trap before the space provided below the elevation of the tile line becomes completely filled.

Sand traps are usually square or rectangular structures located in the tile line. They should have ample storage space below the grade line for collecting and storing sand which is deposited in the relatively calm or slow-moving water in the box. If water moves too rapidly, it may be necessary to install baffles on the floor of the trap to accomplish deposition of material suspended in the water.

When constructing a sand trap, care must be used to prevent "short circuits". In other words, intakes and outlets must not be placed too closely together as, for instance, the inlet in the end of the structure and the outlet so close on an adjoining side that the discharge water from the inlet is projected into the outlet without appreciable slowing of velocity and correspondingly no opportunity given for settling. Such an arrangement interferes with proper disposition of the sand in the trap.

Manholes of sufficient size should always be placed in the top of the trap. At times, such structures are built with an inadequate opening provided for cleaning of the box and drain line.

Often it is necessary to change the direction of the flow of drainage lines. There is no better place to accomplish this than through a junction box or a sand trap. In fact, it is desirable to install a sand trap wherever an acute change of direction must be made.

The work of malaria control drainage under emergency conditions will

probably consist of drainage effected in the shortest possible time, possibly with crude native implements, using unskilled labor -- probably native -- where precision methods such as are associated with the tile drainage will be difficult of realization. In other words, open ditches, and most frequently unlined, will almost certainly be the type most commonly used.

Grades should be established with a surveyor's level, but if one is not available, or if its operation is not understood, then a simple line or string level may be used. These levels are not usually highly accurate, but are of use in establishing crude preliminary lines. In using this procedure the level is suspended on a string, pulled taut between two grade stakes, placed usually not more than twenty-feet apart. The level should be placed on the string about one-third of the distance between stakes and one end of the string elevated or depressed until the instrument indicates that the string is level. Marks are then made on the stake corresponding to the elevation of the string at the two stakes. These two points are then at the same elevation. Differences in elevation are calculated entirely by means of some measuring device such as a foot rule or a tape and the difference in elevation, indicating the chosen gradient, marked on the grade stake. The successful use of this level depends entirely upon the skill and accuracy of the manipulator.

Absolutely flat open ditches will often drain away or concentrate large bodies of standing water, and, accordingly, appear to justify the existence of a ditch without grade. This, of course, is usually not considered good engineering practice, and has the special disadvantage that such a drain will become filled with silt very easily, and in addition, mosquito larvae may breed in such a ditch. There is no advantage to be gained in draining a small area if another breeding area is created in the ditch used for draining. One exception to this rule exists; namely, if the constant level flat drain, installed at the same elevation, is to be used as a sump to remove water from an area difficult of access for larviciding and concentrate breeding in an area where larviciding, or the use of predators, can be more easily performed. Also, construction of such a sump might prevent breeding by changing the area where water remains from sunny to shade or vice-versa.

Open ditches are usually constructed with V-shape and U-shape bottoms. The width, which determines in large measure the carrying capacity is determined by the amount of rainfall, the watershed, the type of soil, and on the general topography.

When excavating a ditch exclusively for malaria drainage, it is well to maintain the bottom width at a minimum in order to concentrate residual waters. "Velocity" or "flow" under these circumstances will be better maintained even when "volume" of water is low. If it is certain that the ditch must carry large volumes of water at flood times, and only small volumes at water periods, it may be advantageous to open a cuneate type drain. This is merely a small subditch in the bottom of a large ditch for carrying off low

volumes of residual water.

Depth of ditches will depend largely upon the necessities of drainage, elevation of the outfall, the terrain through which the ditch passes, and upon the gradient which must be given.

Side slopes are important on any open ditch construction. The slope selected depends mainly upon the stability of the ground through which the ditch passes. If the soil is loose, open sand, then side slopes of as much as two to one (two feet horizontal to one foot vertical) may be necessary to avoid slumping of the sides. If the soil is heavy, as in clay, a slope of one-half foot horizontal to one foot vertical may be permissible. The final top width will depend entirely upon the established bottom width plus the necessary side slope.

Spoil banks are the excavated material. In malaria control, it is good practice to level off these spoil banks at some distance away from the ditch. If the nature of terrain does not permit of such disposition or scarcity of labor contra-indicates this practice, spoil banks should be arranged in staggered formation to prevent the damming back of water. If spreading is impossible, a berm of at least three feet should be left. The berm is the distance between the top of the side slope and the spoil bank. A wide berm facilitates quick maintenance, which is often necessary in malaria control. It also affords a path for the operation of a larvicide gang in case larviciding is necessary, as well as for general inspection of the ditch. Spoil banks should never be left in such condition that they may dam back the flow of surface water into the drain line, and they should be placed on the side of the ditch having the lower elevation.

As indicated earlier, maintenance is the principal weak point associated with open ditch systems. In order to reduce maintenance, the following points should be remembered: (1) good ditch alignment, (2) clean right-of-way, (3) avoidance of excessive grades in unsuitable or unstable ground, and (4) selection of uniform grades which pass from low grades to higher grades rather than the reverse.

Of the special subjects mentioned previously with which the engineer will deal in the works of malaria control, drainage has already been covered in a general way. Fill, impoundment of water areas, and tide gates are covered in special sections later.

Utilization of chemical or organic change in breeding waters is a relatively simple procedure. With preference for breeding waters preferred by individual species established, these waters, at times, can be altered to render them unfavorable for breeding.

It has already been suggested that waters be altered as indicated in accordance with the degree of salt contained therein. Utilization of exposure to sunshine or shade in influencing breeding has been mentioned. In

addition, chemical wastes from factories may serve to render a water unfit for breeding. When sanitary precautions do not preclude the possibility of its use, simple pollution with sewage may decrease or completely curtail anopheline breeding. Naturally, this method would be limited in its application as water polluted in this manner would generally constitute a health menace of another nature in the community where used.

Flushing can be used under certain given conditions. When breeding occurs in the lower reaches of a stream or drainage outfall line, and where facilities are offered at higher elevations for impoundment of waters, these waters at higher elevation can be dammed back and when released, at intervals indicated by breeding, will flush the lower portion of the system, and eliminate large proportions of larvae previously found breeding in the area.

Agitation of water involves artificial production of waves, the wave-action being inimical to breeding. Any device which will produce adequate wave-action can be utilized for this purpose. A water wheel, for example, especially where adequate water power for its function is present, can serve to agitate an impoundment area of limited size to a degree where breeding can be reduced or even eliminated.

Retention reservoirs are frequently utilized, strategically located, to prevent flood waters from causing residual water breeding areas. These waters, after retention, can gradually be diverted into the drainage system, and unnecessary flood of potential breeding land can be controlled. Retention reservoirs, at times, may serve the dual role of preventing flooding, as well as serve for flushing of an outlet system which may breed anophelines.

Manually operated gates are frequently used on retention reservoirs. They are also used on impoundments, where fluctuation of water levels is to be utilized as a means of control. They are further employed to retain impoundments of water caused by high-tides, usually associated with retention of salt waters to produce salinification in an area to a degree where breeding will be eliminated.

Diking is most frequently employed in association with the use of tide gates. If, and when, the elevation of the shore line is so low that high tides overflow a large extent of the shore line, and flood the low lying inland area, then a dike or sea wall should be constructed to prevent these flood waters from entering the low lying inland areas. Openings for points of discharge of inland drainage waters are provided at selected indicated locations in the wall. Tide gates, installed at these discharge points, can then perform their dual function of preventing tidal flood waters inundating the area, at the same time, permitting accumulated drainage waters to be discharged at periods of low tide.

Surface grading is a simple procedure frequently employed to conduct waters in the basin of a drainage system into the central or subsidiary drain-

age channels of that system so preventing the accumulation of residual surface waters. This procedure will be used most frequently where soils, especially, superficial soils, are relatively impermeable.

Surface grading is further resorted to in the preparation of areas if impoundment for fluctuation is to be utilized. The reason for its use is obvious: To prevent the impoundment of small water areas in the surrounding basin of the impoundment as draw-down of the water elevation is being effected for the control of breeding.

It cannot be over-emphasized that the general and natural interest of the average engineer with relation to drainage of waters consists of the removal of flood waters, and the equalization of hydrostatic pressure, particularly, with relation to this pressure on the two sides of embankments or causeways. Graphic demonstration of this practice of equalization of pressure, without thought of malaria drainage, is frequently seen in the high elevation of culverts under roadways and railroads where water areas and resulting lateral pressure exists against the embankment. The culvert at high elevation provides adequate equalization of excess hydrostatic pressure between the two sides, but may leave impounded residual waters which accumulate below the elevation of the misplaced culvert.

To terminate this brief outline of the role of the engineer in malaria control it is indicated to mention again that this role deals largely with the removal and care of residual waters capable of breeding as well as the lowering of ground water table elevations, in saturated and semi-saturated soils, where, because of this high elevation, surface waters exist and breed anophelines.

Routine Malaria Survey

Procedures and Analysis

(In contradistinction to emergency procedures outlined in preceding sections)

Malaria surveys and their analysis yield facts pertinent and indispensable for the most economic and effective application of malaria control procedures. When circumstances permit, they should always be performed. It would be a rare instance when lasting control could be effectively realized without the knowledge acquired through the practice of at least some procedures of the survey.

TO UNDERSTAND MALARIA SURVEYS, THEIR ANALYSIS,  
THEIR USE AND APPLICATION TO FIELD PROBLEMS,  
IS TO UNDERSTAND THE CONTROL OF THE DISEASE.

## MALARIA SURVEY

### 1. INTRODUCTION

A malaria survey is basically a quantitative as well as qualitative measurement. It is directed toward securing information regarding the fundamental factors involved in the propagation of the disease at its source; namely, the anopheline mosquitoes and the human and vector reservoir of parasites.

The survey should serve as a blue-print for the guidance of the malaria department, and should also provide an original baseline from which all subsequent measurements of the course of the disease can be measured.

### 2. REQUISITES

#### a. MATERIALS AND EQUIPMENT

A list of the necessary equipment will not be included here because this subject has been covered in detail by such writers as Boyd (1930) and Christophers, et al, (1936). The reader is referred to the above publications, and others on the same subject. (Also, see special list in this syllabus, page 149, "Clinical Investigative Equipment", and page 147, "Entomological Equipment").

#### b. STAFF

The staff will naturally depend upon the size of the program and the period over which it is to function, but, generally speaking, the essential personnel for any formal investigation should consist of:

- (1) A malariologist.
- (2) An entomologist with special training in culicidology and the use of larvicides.
- (3) A sanitary engineer with special training in hydraulics and hydrography.

### 3. PROCEDURES AND CONCEPTS OF EPIDEMIOLOGY

The survey involves studies covering three different branches of activities; namely, the clinical, the entomological and the engineering. The clinical studies indicate the "what" and "where" of malaria, and, to a certain extent, the "why". Clinical investigations demonstrate what the incidence of malaria is; in what phase the disease exists, epidemic or varying degrees of endemicity; what parasites are involved; and what groups of the populace are harboring the disease. The clinical findings also demonstrate where geographically the cases are located; where cases with parasitemia and gametocytes reside, where breeding areas are likely to be present as based on the geographic location of concentration of cases.

Clinical studies, finally, demonstrate a certain degree of the "why" of malaria in demonstrating the presence of and location of gametocyte carriers.

The entomological portion of the survey deals in large measure with the "why" of the presence of the disease. The "why" of malaria consists largely of the parasite and the vector. The only places the human malaria parasite is known to exist is in the body of man and in the anopheline vector. Sterilize all humans and infected anophelines of the parasite and human malaria would presumably be removed from the earth; a task certainly, at present, impossible of realization. Other palliative procedures based on concepts more practical at the moment must be applied for the control of the disease. It is, of course, conceivable that medication, or other procedure, may some day accomplish that revolutionary and much desired end.

The engineering portion of the survey deals mainly with the "how" to eliminate or control water areas which are breeding or capable of breeding the vectors.

Insect borne human diseases result from the inner-relationship of at least three living entities, the vector, the parasite, and the human individual infected. Disease, though unfortunate for the host, is a natural phenomenon, and malaria is decidedly no exception. The female anopheline is simply performing a function of her life when she secures a blood meal for the purposes of propagating her species, and happens, in the passing, incidentally to secure that meal from man and transmit the plasmodium of malaria.

By a realization of the natural phenomenon of disease a better understanding of epidemiology is born and a more realistic approach to the study and control of the disease is realized.

#### 4. METHODS

A malaria survey should provide the following comprehensive information:

##### a. CLINICAL DATA

- (1) Information on total population; age, sex, race, etc.
- (2) Presence of malaria, its intensity and local distribution. History of disease in locality. Totals of splenic and parasite positives, also classified by race, age, sex, occupation, etc.
- (3) Status in which the disease exists, epidemic or endemic, degree of prevailing endemicity, and general information regarding endemicity.
- (4) Geographical distribution of malaria.

- (5) Recent transmission rate.
- (6) Seasonal variation of the disease and cyclical appearance and disappearance. Occupational status with reference to the disease.
- (7) Species of parasites present, their density, also presence of gametocytes, and relation of parasite positives to splenomegaly.
- (8) Accessory data; economic situation of populace, housing, food, migratory habits of population, medicines used.

b. ENTOMOLOGICAL DATA

- (1) Local species of Anopheles present.
- (2) Absolute and relative density of adults of different species. Seasonal variations in their presence and densities.
- (3) Determination of the anthropophilic and zoophilic tendencies of the species.
- (4) Identification of the local vectors of the malaria parasites.
- (5) Determination of the varying types of water areas where vectors can breed and specific localities which produce vectors, and their relationship to areas to be protected.
- (6) The ecology and bionomics of anopheline vectors (inclusive of flight range) with a view to the application of control measures based on their habits, and also habits of domesticity.
- (7) Presumptive location for capture stations and traps.

c. ENGINEERING DATA

The detailed studies of the engineers in determining the best methods for the treatment of waters should be dependent upon previously secured entomological findings, particularly the presence of actual breeding of the vector and the flight range.

The engineering studies should supply:

- (1) General maps of the locality for engineering, clinical, and entomological purposes.
- (2) Notations on maps of the general topography of the locality with water courses and water sheds marked as well as the logical discharge points for water.

- (3) Special topography of projected drainage lines.
- (4) Type of soil involved in each projected drainage system and its permeability.
- (5) Notation of water areas potential for breeding.
- (6) Indicated meteorological information.

## 5. THE CLINICAL SURVEY

### a. SPLENIC EXAMINATION

The splenic examination, with associated blood examinations, constitutes the basic activity of the clinical survey.

### b. SAMPLE FOR EXAMINATION

The school affords a focal point where the younger age groups of both sexes, various races, and representatives of differing social status congregate. Therefore, it is a convenient place to conduct the major investigations of the clinical survey. However, on occasions, other age groups, including infants and adults, must be included.

The clinical survey should, when possible, be made at the period of the year of greatest intensity of the disease, or immediately following that period. Once this period has been ascertained, on the basis of history, statistical data, or previous surveys, the examinations should be repeated, when indicated, at the same period of each year in order that the results may be comparable. If two surveys a year are possible, an additional one can be made prior to the malaria transmission season. The findings in this survey would represent largely residual malaria from the previous transmission season.

### c. GENERAL PROCEDURE

Splenic examination is made preferably with the patient in a recumbent position, with knees flexed, and with abdomen bared. The examiner palpates lightly with the right hand held more or less at right angle to the costal margin. Light pressure by the examiner, with the hand flat upon the abdomen, and relaxation of the patient, associated with full, deep, abdominal breathing are essential requisites for successful examination. Thoracic breathing, with tense muscles, can completely conceal the existence of some enlarged spleens which otherwise might be readily palpated below the costal margin.

The record of results should take the form of a statement of the relative position of the lower border of the enlarged spleen with reference to the costal margin. For this purpose, Boyd's classification of spleens has been found practical. His classification divides enlarged

spleens into five groups, namely: Palpable on deep inspiration (P.D.I.), and Numbers 1, 2, 3, and 4, the progressive numbers indicating progressive degrees of enlargement, varying from Number 1 at the costal margin on normal inspiration to Number 4, which extends below the umbilicus. Number 2 enlargement may extend half-way to the umbilicus, and Number 3 from the former limit to the umbilicus. It has recently been advocated with considerable justification that splenic enlargement be indicated by the Numbers 1, 2, 3, 4, and 5, in which situation the P.D.I. spleen would correspond to the Number 1 in this classification, and Numbers 1, 2, 3, and 4 would become Numbers 2, 3, 4, and 5.

A practice which has been found to be very helpful in surveys is to examine for splenomegaly the population selected as a sample, taking blood smears from each person with splenic enlargement. In association with this, blood smears are taken from a certain proportion (every second, third, etc.) of those persons resulting negative to splenic examination. An analysis of the results secured from the examination of these smears assists in definitely confirming the degree to which the splenomegaly is indicative of active malaria infection, but does not constitute a parasite index. Other observers prefer a blood examination of all persons in the sample, from which to compute the conventional parasite index. Such procedure is, of course, most time-consuming. It should be remembered that at certain periods in the approximate seven-year cyclic disappearance of clinical manifestation, especially in areas where a moderate endemicity is not exceeded, it is difficult to demonstrate at all times the presence of parasites even with the examination of thick blood smears.

If malaria be present, the sample of the population subjected to splenic examination will include a varying proportion of acute clinical cases and latent or chronic infections. Those with detectable enlargements may include a few persons already recently recovered from the disease. On the other hand, a few whose blood examinations are positive will not exhibit detectable splenomegaly. These possibly represent very recent infections.

Blood and spleen examinations should be made and used, for more comprehensive results, in conjunction with each other.

#### d. BLOOD EXAMINATIONS (General Procedures)

A satisfactory manner of taking blood specimens is the method prescribed by either Barber (1929) or Christophers (1936). The thick smears are placed at one end of the slide, and a thin film on the other. When dry, the identifying number or name can be written in the thin film with an ordinary pencil.

A certain minimal time interval should be devoted to the examination of a thick film before concluding that parasites are absent, as judged by routine laboratory procedure. Barber (1929) suggests that this limit must

vary according to the expertness of the individual examiner. A minimum of five minutes for examination of thick smears is considered good practice.

When blood smears are indicated, these should always be taken at the same time splenic examinations are made. Fresh smears must under all circumstances be protected from insects, especially flies. If impossible to examine immediately, the slides should be stained and preserved for a later date.

#### e. RECORD CARDS

Record cards with peripheral perforations, to be punched for positive observations, facilitates the recording of all data and subsequent analyses.

For permanent records, the minimal general information gathered from cases examined for splenic enlargement or by blood smear examination should include date, locality of examination, name, age, sex, race, residence, and length of residence in house and locality. On each case should be recorded, as well, the result of splenic and blood examinations.

If analysis facilities permit, additional information relating to the following may be secured:

- (1) Species of parasite, gametocytes.
- (2) Occupation.
- (3) Malaria medication taken.
- (4) Condition of dwelling and protection against mosquitoes.  
(Difficult to secure without a domiciliary visit).
- (5) Attacks of fever over a period of the last year.
- (6) Homoglobin percentage, if desired.

#### 6. THE ENTOMOLOGICAL SURVEY

A mosquito breeding area is not a chance occurrence. There is a distinct relationship between that area and the source of a blood meal within flight range of the mosquito in question. There is no reason to believe that mosquitoes would, under ordinary circumstances, (except perhaps for hibernation or aestivation purposes), lengthen the flight necessary to secure a blood meal. With the possible exception of some salt marsh mosquitoes, it can be said that, when a breeding area is firmly established, it is clear evidence that a source of blood meal is readily accessible, be it animal, bird, or man. A breeding area of an anopheline vector of malaria spells potential malaria in that district if humans with parasites move into that area and are substituted for the local blood meal previously enjoyed by the existent anopheline fauna.

Once again the propagation of the disease malaria is influenced by natural phenomena, in this case, the natural life habits of the vector.

Entomological investigations in a survey are customarily limited to the area to be controlled, including, usually, as based on flight range, an additional radius of one mile around the actual area. These investigations include field observations on the larval and adult stages. The ova, at times, are also included, but these observations, as a rule, are not highly essential to the practical ends of a survey. In order to secure information on seasonal and yearly variations, primary entomological studies are preferably continued for a year or more, when possible. However, the exigencies of practical control work will rarely permit this.

Location on a spot map of the residence of the infected persons, as revealed through splenic and blood examinations, affords a real and practical lead to the anopheline production areas, at least those of major significance, producing vectors. However, all water areas must be studied which are within average flight range of the district in which control is to be instituted. Actual investigation primarily includes the capture of adults by means of capture stations and traps and the larval search for breeding areas.

If early in the season, and breeding is just beginning, search for larvae alone is most practicable; whereas, if the season is more advanced, both lines of investigation should be performed simultaneously. It should always be remembered, however, that adults are the most important element. They actually bite and so transmit the disease.

The field observer must be painstaking as his investigation constitutes a sine qua non in formulating plans for control. He should become acquainted with the local terrain and adjacent regions. Topographical maps can assist in the localization of potential breeding places.

Entomological investigations are performed largely with the aid of a flashlight and catching tubes (either suction or chloroform), dipper and wide mouthed pipettes, rubber boots, labelled specimen boxes and bottles for individual collections, and a determination to discover mosquitoes if and when present. A notebook for recording the date, locality and time spent in capture is also required.

All observations should be entered on permanent record cards. The records should afford the following minimal data:

- (1) Exact location of survey and date when performed.
- (2) Type of water areas, and general characteristics of water.
- (3) Exposure to sun or shade.
- (4) Associated meteorological data when available, especially

rainfall and winds, and seasonal variations of these.

- (5) General information on vegetation in water area.
- (6) Number of larvae found, in accordance with the adopted unit of measure, and their instars and identifications.
- (7) Pupal shells.
- (8) General idea of distribution of breeding and its density (per square meter, for instance, or per dip).
- (9) Distance to possible source of blood meal of adults.
- (10) Distance of breeding area from area to be protected.

All actual and potential breeding places should be represented on maps by conventional signs distinguishing the actual from the potential. Note: The presumptive permanency of water remaining in each area should be recorded.

Most specifically, the entomological survey should reveal where the proven vector species breed, the type of area in which breeding occurs, and the districts that adults frequent. Observations on adults are secured from nocturnal catches, the use of traps, and captures in diurnal shelters. Such places, if routinely visited, are called capture stations.

Capture stations are localities chosen for capture and should represent places where anophelines congregate. They may or may not be related to a source of a blood meal. A latrine, a shed, in or under a house, stable, in a culvert, or any protected locality affording shade and humidity, and protection from winds, may be selected to serve as a capture station. A trap is a device for attracting and retaining anophelines (partially at least), and serves as an additional means of sampling the mosquito fauna.

All collections from each location should be kept as separate units in appropriately labelled boxes for identification and subsequent study.

As control work progresses, routine entomological investigations afford a check on the progress of the control. Permanent capture stations should be chosen for routine visits. These should be selected only after preliminary investigations have determined that the sites are actually frequented by adults. They should represent a routine meshwork coverage of an area designed to secure complete entomological information, both positive and negative. Traps should also be located with definite relation to known breeding areas, as well as the area to be protected, and in such manner that they cover the total area to be studied in a comprehensive manner. Such stations should be visited at weekly or ten-day intervals.

The importance of certain breeding areas with reference to their

actual role in producing anophelines responsible for malaria transmission in an area, particularly if located at the maximum flight range of a species, can be demonstrated by the recovery in special catching stations of stained adults released for this purpose. Another procedure which can assist in such study is the location of catching stations located from each other at a more or less constant distance apart between the breeding area and the area to be controlled. The density of captures in these stations, at ever increasing distance from the breeding area, can give valuable information on the source of breeding.

Some expression of the density of anophelines should be adopted. When density is not great, comparison may be made between total catches, but when more abundant, results may be expressed as the number caught per man-hour (Boyd, 1930), or by careful observance, the total number may be estimated. At times, collections are repeated from the same surface area, and successive results are then felt to be comparable.

The permanent records on adult captures should list the following points:

- (1) Places, date and hour.
- (2) Type of station, diurnal or nocturnal catch.
- (3) Weather conditions.
- (4) Time spent in capture.
- (5) Numbers of each species identified; also, numbers of males and females.
- (6) Protection from sunshine and winds.
- (7) Humidity.
- (8) Source of blood meal.
- (9) Prevailing winds.
- (10) If traps be used, factors of fundamental importance associated with capture in these traps should especially be recorded; namely, humidity of the area and the trap's exposure to or protection from wind and sun, its proximity to a blood meal, and distance from a productive breeding area.

Analysis of the data should yield the following information:

- (1) Species present.
- (2) Their local density and distribution.

- (3) Seasonal variation.
- (4) Their relation, seasonal and geographic, to presence of clinical cases and to anopheline production areas.
- (5) Flight range and direction of ingress or infiltration.
- (6) Observed points in Bionomics and Ecology.

The data should be entered on the entomological maps.

a. THE VECTORS OF THE LOCALITY

It is not necessary as a rule to make specific appraisal of the actual or potential role of the local anophelines as vectors in every small locality of the same general district. The results of intensive studies performed in a few typical and sample localities will likely afford significant information applicable to wide areas.

If indicated, pertinent information relative to feeding habits of female adults may be obtained from the results of precipitin tests performed on a series of bloods removed from the stomachs of freshly blooded anophelines. Such studies identify the host from which the blood meal was taken. If the presumptive evidence suggests a species may possess anthropophilic proclivities, its susceptibility to infection with malaria parasites, if unknown, should be determined. The infection of this species may be demonstrated in anophelines caught under natural conditions. For technique of the precipitin test, see Boyd (1930).

7. ENGINEERING SURVEY

If topography is unfamiliar or maps unavailable, maps must be made immediately for the use of the clinical, entomological, and the engineering phases of the program. Hulse's description (1922) of sketch mapping is very satisfactory as an introduction to the practical preparation of work maps. In entering data on these maps, attention must be given to:

- (1) All residences and buildings and their distinguishing identifications.
- (2) Water courses and collections of water, areas of low elevation which may hold water temporarily, and their location with reference to areas to be controlled.
- (3) Information relating to the following may also be required, viz:
  - (a) Geological formation of the region, types of soils.
  - (b) Past and present record of precipitation.
  - (c) Average yearly amount of rainfall.

- (d) Average, maximum, and minimum humidity.
- (4) Possible practical, terminal discharge points for water sheds.

The detail of the engineering survey will naturally be dependent on the type of control which is selected for the locality. If personally applied measures are to be used (repellents, nets, aerosol sprays, medication), no engineering survey will be necessary as alteration of the environment responsible for production of the vector is not to be attempted. Once it is decided that control of breeding area is to be attempted, unless this control be larvicidal, the engineers must make detailed studies. Depending upon the method chosen, these studies would involve such subjects as the following:

Drainage, (open, closed, lined, unlined, intercepting, surface), fill, alteration of water content by salt or pollution, impoundment and fluctuation of water levels, flushing, retention reservoirs, use of tide gates, agitation of water, influencing of hydraulic factors of water currents, and change of environment for breeding from the ecological standpoint.

Probably the procedure which would have the widest application of the measures noted above would be that of drainage to remove indicated breeding waters. Accordingly, the studies indicated in that phase of engineering activities will be outlined.

Once a water area has been designated as responsible for breeding anophelines, which are actual agents for the propagation of the disease in the locality, and once drainage be selected as the remedy for that situation, certain engineering studies must be performed before embarking upon the drainage project. Such studies include:

- (1) Topography of the area; general, if maps are available; detailed, if map-making is necessary.
- (2) Water shed.
- (3) Volume of water to be removed.
- (4) Rainfall data.
- (5) Logical and effective discharge point.
- (6) Subsurface water level.
- (7) Types of drainage system to be installed (surface drainage, open conduits, closed conduits, lined or unlined, number of main drains, and laterals, intercepting systems).

- (8) Location of central drainage system.
- (9) Location and elevation of outlet.
- (10) Gradient of drainage lines.
- (11) Amount of cuts necessary and quantity of earth to be removed.
- (12) Waters which will remain undrainable by system because of their elevation lower than the elevation of the drainage line.
- (13) Amount of fill necessary for these areas.
- (14) Permeability of soil.

## 8. ANALYSIS OF MALARIA SURVEY DATA

Malaria surveys and their analysis are most practical procedures as the guiding agent for practical control procedures. If it be demonstrated by the survey that a certain occupational group of people are those acquiring malaria in the populace under question, control measures can be directed toward the correction of the epidemiological factors which surround this group to the large exclusion of other groups. If it be demonstrated that certain age groups constitute the seed bed of parasites and gametocytes, special measures to limit the spread of the disease can be directed toward that group, and not toward the general populace. If the survey demonstrates that P. falciparum is responsible for the major portion of malaria in the group studied, the malariologist knows immediately that he must be on the alert for cases of severe clinical manifestations and even mortal results.

Knowledge of the vector revealed by the survey is of utmost importance. If, for example, the vector be A. gambiae, a highly domesticated anopheline, the problem of transmission and control is greatly altered from the situation where A. bellator is responsible for conveying the infection. If the vector proves to be of the species that breeds in sunlight, clearing of forest areas where water is present should be performed with the full knowledge that additional breeding area may be formed.

For all who have worked in malaria control, it is clear that the malaria survey and its analysis is not academic. It is most practical and, with existent knowledge, still constitutes the keystone of the arch on which well directed control measures rest.

### a. DETAILS OF ANALYSIS

The different phases of the survey, clinical and entomological, are complementary to each other, and, in appraising the situation, their results should be considered jointly. An analysis of the survey data should supply the information on the fundamental epidemiological factors which were noted in Section 4, "Methods", Pages 38-40.

History of the locality and already existent statistics may prove of value if they have been sufficiently comprehensive. At the beginning of the survey, they may indicate that period of the year which represents the height of the malaria season, and in areas where infection has been relatively heavy, they may give a general history of the period of years over which malaria has been present, and indicate the cyclical tendency of the disease. Such information should be accorded, however, only the importance that its origin and reliability merit.

In dealing with an analysis of a malaria survey, it should always be remembered that persistent splenomegaly is an evidence of an immunity and parasites a sign of recent activity or recent transmission.

The perforated record cards facilitate the work of analysis. Those pertaining to each geographical unit should be analyzed separately, and the subtotals for each area summed up to render the whole picture. From these cards information is quickly secured regarding:

- (1) Age, sex, and race of those examined.
- (2) Numbers in each of the above groups with splenomegaly.
- (3) Numbers of cases with splenomegaly found harboring parasites.
- (4) Numbers of cases without splenomegaly found harboring parasites.
- (5) Incidence of parasites in each geographical subdivision.

(Note: These results should be spotted on maps, showing the places of residence of those examined, together with essential findings).

#### b. SCOPE OF MALARIA PROBLEMS

The analysis of blood and splenic findings considered separately and together will indicate:

- (1) The local geographic distribution of infected persons. This distribution may be diffuse or concentrated.
- (2) The phase and intensity of malaria, i.e., whether the incidence observed represents an epidemic or endemic prevalence, as well as the degree of the grade of the latter.
- (3) Total percentages found positive, and size of spleen. (Average spleen and average enlarged spleen.

#### c. TYPE AND CHARACTER OF THE MALARIA PROBLEM

The analysis gives evidence on the following subjects:

- (1) Endemic characteristics of the disease (when not hyper-endemic)

As a rule, when active transmission is not present in an endemic area,



the following general conditions will maintain:

- (a) Groups most commonly showing clinical manifestations under these circumstances are children and new arrivals. (Spleen usually hard, not painful). No evidence of general manifestations of a febrile disease among the populace.

- (b) Splenic and blood findings:

Relatively low rate of parasite carriers, as compared to periods of active transmission, in cases found with splenomegaly, and lower rate in cases negative to splenomegaly.

Small percentage of gametocyte carriers among parasite positives.

Percentage of parasite positives, as well as cases with splenomegaly, decrease rapidly with age.

Percent of total population found positive to splenic enlargement dependent upon grade of endemicity present.

- (2) Epidemic status (uncomplicated) shows the following characteristics:

- (a) Little or no past history of malaria in region and no statistical evidence; evidence of presence of an acute febrile disease.
- (b) Spleens soft and frequently painful. (Notations should always be made on this subject in making surveys). Few, if any, large or hard spleens.
- (c) Large percentage of positive bloods among the splenic positives.
- (d) Large proportion of positive bloods among the group negative to splenic examinations.
- (e) As judged by splenomegaly or parasitemia, no particular variation in rate of infection by age, or in new-comers to the community.
- (f) Relatively high rate of gametocyte carriers present in all age groups.

(It should always be remembered that race may have an important bearing on evidence of infection, and the findings in one race should not

necessarily be made applicable to another in the same locality nor applicable to potentiality of malaria in new-comers of a different race. Findings in one age group should not be assumed to be applicable to another group, and the same may be said in certain instances regarding infection rates in persons of differing economic status).

It is of prime importance that the malariologist recognize the status of the disease with which he is dealing, be it epidemic, or varying and increasing degrees of endemicity; namely, mild, moderate, severe, or hyper-endemic. Difficulty of control and time necessary to effect it will depend largely on the status of the disease encountered. A first epidemic in a new locality probably presents the easiest problem for control. In such a locality there would be no large reservoir of parasites in persons able to be up and around transporting their parasites with them and capable of infecting new mosquitoes. Cases of the disease could be easily recognized by clinical symptoms and isolation quickly effected. Infected anophelines on the wing would not necessarily be widely spread throughout the whole district.

In all probability predominant factors favorable for the development of an endemic area do not exist.

On the other hand, endemic situations have developed because nature has decreed it. She has lavishly offered those factors which favor transmission once a parasite has been introduced. Rainfall, humidity, climate, wind, relation of source of blood meal to potential breeding areas will be found in such areas in a state of coalition or fusion favoring the development of an endemic status of the disease. With nature supplying the factors to promote an endemic status of the disease, control will be found to be more difficult. In such an area of severe or hyper-endemicity, there will exist many ambulatory human reservoirs of parasites whose clinical condition does not confine them to bed and who can carry their gametocytes with them everywhere. There will be an efficient vector from the standpoint of its domesticity or perhaps simply by its preponderance of numbers and a correspondingly high percentage of infection on the wing will be almost constantly present.

On the other hand, a new epidemic area usually develops because of factors which have been altered, the introduction of a parasite, the introduction of a vector, or changed conditions, frequently man-made, which favor the production of a vector already biologically present in a locality, but not present to the degree of important economic disease production. The malariologist must know this problem. To know the status in which the disease exists is of the highest import in foretelling the difficulties to be encountered in control, and in preparation for material and personnel in the work of control.

#### d. ROLE OF OCCUPATION IN INFECTION

Occupation may predispose toward or protect from infection. The effect

of occupation will be revealed by analysis. Age and sex, with their different customs and habits, may produce the same result. For example, occupations involving night work in unprotected buildings and favoring one sex or specific age group may predispose to infection. If unrecognized, such considerations may be misleading in the analysis of the district as a whole, but if recognized, can be most useful when control measures are being weighed and control being instituted. Sex, or even age, may predispose toward certain types of employment or activities which expose to or protect from malaria.

#### e. SEASONAL VARIATIONS: ITS IMPORTANCE IN ANALYSIS

If sufficiently accurate, history and statistical records can help in this field. More definite data, however, are secured through continued survey and by correlation of clinical cases with the degree of anopheline density.

A knowledge of seasonal variation is important in controlling seasonal malaria.

#### f. PLASMODIA

The presence of parasites and the species of plasmodium present, whether Plasmodium vivax, malariae, or falciparum, indicates:

- (1) Degree of seriousness of sudden, severe clinical course of disease when P. falciparum is present.
- (2) Possibility of high transmission rate with P. falciparum.
- (3) Possibility of suppressive medication, 0.1 gram daily of atabrine, serving as therapeutic cure with P. falciparum if continued for one month after exposure to new infection has passed.
- (4) Tendency to relapses with P. vivax.
- (5) Recent transmission when any species of parasite is found in infants, or new-comers to the community. (Transmission occurred within the life of infant or time of residence of new-comer).
- (6) Predisposition for immediate transmission in area under consideration when gametocytes are present.
- (7) Recent activity when high densities of parasites are encountered per field in microscopic diagnosis.
- (8) Probably epidemic phase of disease when parasites are equally

present in all population irrespective of age groups. (Especially when large, hard spleens are not prevalent).

#### g. GENERALIZATION ON ANALYSIS OF SURVEY

As stated above, splenomegaly, when not caused by other conditions than malaria is a sign post of the body's attempt to produce immunity, and parasites indicate recent transmission. The knowledge of these facts and their inter-related manifestations represent the handy tool, the "open sesame", of the analysis and understanding of malaria surveys.

Spleens, attributable to malaria, when found enlarged, paint the recent and even the past history of the locality from the malaria production standpoint, and indicate the potentiality of that district to harbor and produce the disease. The presence of parasites, and concomitantly gametocytes, is the red light signal that transmission has recently been present and, in the presence of the vector, can again become active. The splenic and blood findings then taken together depict past history and future potentiality of this area, as well as its immediate proclivity for transmission.

The finding of hard, unpainful, enlarged spleens, attributable to malaria, is the first indication of an objective nature that the area is an endemic one. This original lead would be substantiated later in the analysis by finding a larger number of cases of splenomegaly in young age groups or in new-comers to the community than in the older age groups or long residents. The actual total numbers of persons with splenomegaly theoretically bear no relation to the diagnosis of endemicity or epidemicity as, theoretically, to mention an extreme supposition, 100% of persons could have enlarged spleens in an epidemic area as well as in a hyper-endemic one. The character of the spleens, however, would be different.

In extreme hyper-endemic areas, the decrease of splenomegaly, as older age groups are involved, will be less apparent than in areas of lesser degree of endemicity since, as 100% splenomegaly is approached, practically every one, old and young, new-comers and infants, have the signs of the disease, including an enlarged spleen. Under such conditions, the presence of hard, unpainful spleens, the large number of large spleens, especially in older age groups, the decrease of clinical manifestations in older age groups, and finally, the high percentage of all the populace showing splenomegaly, would be the cardinal points on which the status of hyper-endemicity would be established.

During periods of active transmission of the parasite in hyper-endemic areas, the percentage of persons demonstrated with the parasite, as well as the density of parasites, will be found in most instances to be increased.

Such a situation would represent the presence of active transmission, or stated differently, an epidemic flare-up of the disease super-imposed on a basic endemic situation.

Splenomegaly, character of the enlarged spleen, parasites, their density, differences in clinical signs as manifested between age groups and between new-comers and long residents of the community, the inter-relationship of those with splenomegaly found positive, as well, to parasites; these are the common criteria on which a diagnosis of the status of the disease is made.

#### h. TRANSMISSION

This situation is indicated by the rate of infection in infants, in immigrants, and in non-immunes, and by percentages of cases with parasites found in whole population, as well as high percentages with parasites in splenic positives, and by the heavy density of parasites per individual infected.

Transmission is a function of presence of gametocyte carriers, the efficiency and density of the vector involved, and presence or absence of immunity in the subject.

Some special features of the vector in relation to transmission are enumerated below.

- (1) The entomological survey demonstrating the vector or vectors and the areas responsible for breeding indicates species sanitation where this can be applied.
- (2) Larval observation associated with adult captures indicates whether concentration of clinical cases corresponds to known production areas. If clinical cases exist where no production areas are recognized, these production areas must be sought. Moreover, if adult captures indicate that a breeding area is near, but has not been located, it must be discovered.
- (3) Seasonal density of Anopheles suggests the period of the probable height of the clinical malaria season.
- (4) Knowledge of the bionomics of the vector species may suggest the adoption of a method of biological control.
- (5) Presence of males indicates a breeding area is not distant and young females demonstrates breeding is in progress, or of very recent origin.
- (6) Meteorological studies show the relationship of Anopheles production to these fundamental conditions.
- (7) Conditions of ovaries indicate age of females. Many gravid females indicate water not available for oviposition.
- (8) Deposition of fat in body of females indicates hibernation

present or imminent.

- (9) Flights in excess of normal flights usually indicate hibernation.

With all the facts of the survey and its analysis at hand a practical strategy of control can be contemplated.

## 9. CONCEPTS OF CONTROL

The survey and its analysis places the key to the malaria control program in the hands of the malaria control personnel and upon their judgment rests the ultimate solution.

### a. GENERAL CONSIDERATIONS

Under conditions prevalent in peace times, many conditions will govern the selection of control measures from those available. Under normal conditions there are special indications for the employment of each measure and the entire situation should be carefully appraised. Under normal conditions there is no more justification for the recommendation of utilization of all methods in a given situation than there is for writing of "shot-gun" prescriptions, an old clinical practice now generally discredited. However, special emergency conditions may alter this situation, and make these "shot-gun" prescriptions not only indicated, but mandatory.

Experience of recent years and new developments and discoveries are altering the complexion of malaria control programs. In the future, it may be that the old stand-by procedures of control will be supplanted by newer and different methods. The previous concept of stable, sound methods was based largely on control of the environment by elimination of the mosquito through draining, filling or larviciding breeding areas or by mosquito-proofing to prevent infected anophelines from biting uninfected man and to prevent uninfected anophelines from acquiring the infection from biting infected man. These procedures are admittedly cumbersome and costly, but, adequately applied, they work.

Drainage, fill, and larviciding represent costly measures. They are directed toward eliminating or greatly reducing the anopheline population responsible for transmission of infection.

It is safe to assume, however, that many of these anophelines thus eliminated at considerable cost would have been side-tracked in their search for a blood meal by sources of blood other than man, and, accordingly, would never have transmitted malaria anyway.

New discoveries and adopted procedures have suggested new concepts of approach to malaria control. Only time will prove their effectiveness. Spray killing solutions of an aerosol nature and use of residual DDT in houses have opened new lines of thought. These measures can be applied in

buildings, houses, dwellings, offices, schools, where the disease is, in large measure, actually being transmitted. Only those anophelines that actually bite man will ever transmit human malaria. If that group is constantly being killed, malaria will be controlled. Apparently the best single locality to kill that group constantly would be at the spot where that specific group of anophelines continue to come to bite man; namely, in and around places where man congregates, particularly during the hours generally corresponding to dark.

- Spraying inside of buildings which are used at night, with an efficient aerosol spray, will kill numbers of infected anophelines before transmitting or acquiring the infection. Painting or spraying with suitable DDT solution the interiors, exteriors, screens and surrounding associated buildings, which are used at night, will accomplish a similar end against those anophelines, vectors of the disease, which alight upon it for sufficient time. (Note: In an experiment at the Army School of Malariology, it was demonstrated that 100% of A. aegypti which rested on dry DDT crystals for two minutes were dead within two hours after such exposure under laboratory conditions. A similar mortality was found with laboratory reared A. albimanus which were exposed to only four instantaneous contacts with DDT in solution).

It is obvious that aerosol spraying or application of residual DDT on walls of buildings could greatly reduce malaria by concentrating the attack on killing both infected and uninfected anophelines which are in the act of attempting to bite man. Mosquito mortality, under these circumstances, would increase in accordance with increasing degree of domesticity characteristic of the species involved.

Furthermore, new drugs of considerable suppressive efficiency are appearing which involve dosage only once a week. At times, there is reason to believe that this suppressive dosage continued for a sufficient time, when no more infection is being acquired, will cure the infection. Such drugs could theoretically eliminate a parasite from an area.

Accordingly, at the present time, there appears to be a hope justifiable, if future experience supports present trends, that personally applied measures directed toward the individual and his place of abode might replace the more laborious and costly environmental control.

Under these circumstances, a hope, a prophecy, a trial of today may be a fact, a routine procedure of tomorrow. Still, however, the prudent malariologist will outline his stable control program, under non-emergency conditions, in accordance with the proven and accepted facts of the moment.

#### b. TYPES OF CONTROL

From the standpoint of the types of control, there can be various classifications, but one very practical classification is that based on the time over which control is desired; namely, emergency, temporary, semi-

permanent, or permanent. Different procedures fall naturally under these headings.

Emergency control would be advocated for bodies of men or individuals moving through malarious territory or remaining for short periods in such areas. Such measures are especially adapted to armies at war or on maneuvers. The methods used are largely personally applied measures in which malaria discipline plays the leading role. Malaria discipline, in this instance, can be defined as readiness and desire to utilize protective measures based on an understanding of the role of these measures in prevention of the disease. These measures include the correct use of aerosol sprays; protection of the body from bites by clothing and the proper use of repellents; use of mosquito nets; suppressive medication, not only to limit clinical symptoms, but surely to lower the parasite seed bed in man and possibly to cure, at times, the disease; governing of individual activities to the end that undue exposure to infection is not courted in the towns and buildings of an area where malaria is known to exist. For the rational application of such procedures a survey is scarcely necessary. Little more need be known than the fact that malaria has been reported from the area over the past. Of course, entomological findings would be useful, especially a knowledge of the domesticity of the vector or her more sylvan habits and the season of the year, if such exists, when she is not on the wing. It is always of interest to know if *P. falciparum* is present in a locality because of its lethal capacity when unrecognized and treatment inadequate. However, if the suppressive dose of medication administered is adequate, information regarding the parasite is not necessary.

Temporary and semi-permanent control frequently overlaps each other, and it is difficult to draw clear and firm lines of demarcation. Semi-permanent measures of control are indicated purely on the basis of the time element of the prospective length of residence in a locality or based on the evaluation of results produced measured against the effort necessary to establish some environmental control to relieve the necessity and monotony of self applied emergency measures.

Mosquito-proofing of buildings is classified surely as a long lasting semi-permanent measure and can relieve greatly the necessity for incessant application of repellents and spray killing. Mosquito proofing, to be effective, must be well performed and rigidly maintained. Again, no special knowledge, revealed by survey, is necessary to warrant the use of this procedure other than the fact that malaria is known to exist in the area.

Larviciding is classified normally as a temporary measure. However, it can fall in the category of an emergency procedure or even perhaps a permanent measure. Application of larvicides by airplane, under emergency conditions, can precede occupation of an area by troops when the only known facts are that malaria actually or probably exists and presumptive breeding areas can be seen from the air. Its use further could be justified in wartime or in an emergency as a temporary measure where obvious anophe-

line breeding areas are found after entering an area, even though an actual vector is not identified, but when potential vector breeding areas are within easy flight range of the area to be protected. Of course, in such a situation, cost would not be an element in deciding on its use. Protection of the force would be the deciding factor. When used as a semi-permanent procedure, its use should be predicated on entomological knowledge of actual areas breeding the vector, flight range and the interval of time elapsing in the area between the egg and pupal stage of the aquatic cycle of the vector. Under such circumstance, larvicides are usually used as an adjunct to other procedures and should be replaced, when circumstances permit, by temporary or permanent drainage or fill.

Larvicides could conceivably be used as a permanent measure when the cost of really permanent disposition of the waters is out of all proportion to the importance of the program.

It has also been advocated as a permanent procedure when large breeding areas have been sumped to a small surface area where larvicidal application is easy and where final removal of the water presents great difficulty or would be accomplished at excessive cost.

A procedure which would be classified today probably as a temporary or semi-permanent one, because of the need of repetition, is painting or spraying of buildings with DDT solution. In the future this procedure may fall into the classification of permanent if it be proven that the cost is low compared to efficiency of results. Even though needing repetition at intervals, it would then merit this classification of permanent since it could conceivably be the method of choice for the permanent control of the disease.

Based on the proven facts of today permanent control implies, in large measure, the alteration of the environment to render breeding water unfit for breeding. Accordingly, as the expense of such programs is usually large, clinical survey findings must define the clinical type of the problem, the entomological studies, the areas responsible for breeding the vector, and the engineering side must indicate the possible and desirable choice of method for control of breeding areas.

It is a rare occurrence that a malaria control program will rely upon one procedure of control, even though that be drainage. Drainage construction consumes time and much money. Accordingly, accessory measures are frequently employed simultaneously even though it be known that the ultimate project is designed to be as permanent as possible. Under these circumstances, a survey is invaluable, indicating as it does clinically where infected cases with parasites reside, so suggesting the locality and importance of breeding areas, and also locating reservoirs of parasites; whereas, the entomological studies indicate quick measures which may be taken to improve the situation, such as justifiable temporary drainage or larviciding.

### c. FACTORS INTER-RELATED WITH A CONTROL PROGRAM

When a control program is being considered, due weight must be given to the money available and the urge for complete or partial control over a time limit. With the military, in wartime, the slogan is "control the disease". In enterprises of a local community nature in peacetime many considerations must be borne in mind. The survey will indicate the magnitude of the project and from the studies performed estimates of costs can be prepared.

Among considerations of a general and non-technical character influencing the selection of types of control are:

- (1) The extent to which the problem is of public or private interest. This has a bearing on the authorization for and financial support of the work.
- (2) Whether the sponsoring agency, official or private, will support and maintain a long-term policy relating to malaria control.
- (3) The available or potentially available resources which may be devoted to malaria control. While under some circumstances outside support may be secured from official or unofficial sources, in the main, programs must be adapted to the local resources. Since local resources usually have definite limitations, the husbanding of these through economical planning and execution is essential.
- (4) Whether the population affected are relatively dense suburban, or sparse and scattered rural groups, or, on the other hand, stable or migratory.
- (5) Whether the malaria situation is generally recognized as a long-standing endemic, or the emergency arises from either an epidemic or the creation of situations which potentially favor epidemic extension. Situations of the last type may imperil the success of military or commercial undertakings in endemic areas.
- (6) The emergency character of the contemplated control, for example, in the case of temporary military sites or during the progress of maneuvers or war itself.

Assuming that interested agencies are disposed to devote resources to the solution of the malaria problem, consideration may be given regarding the objective of the program, whether the aim is the eradication of the disease or measures directed toward control of malaria in the area. These terms have different implications. The former, "eradication", would signify an intent not only to exterminate the infection from the area, but to

make its subsequent return practically impossible. The latter, "control", implies a practical reduction in the incidence of the disease to such a low level that it no longer constitutes a social or economic incubus on the community. In areas of epidemicity, or of moderate endemicity, eradication may be a practical objective. In most areas of higher endemicity, where the existent phase of the disease has proven the presence of factors favorable for its propagation, it will probably be necessary to seek simply a reduction in incidence.

Furthermore, in the present stage of our knowledge, consideration must be given to the question whether the outlay for the contemplated program should be largely one of capital investment for permanent or semi-permanent work, which will minimize maintenance or recurring expenditures, or whether it will be devoted to measures which, while initially cheaper, will require constantly recurring expenditures in order to conserve the benefits acquired. The former policy is, in the interest of genuine economy and based on our present day proven knowledge, best adapted to areas of stable populations of relatively high density, the latter to emergencies or situations regarded as temporary, or possibly to sparsely settled rural areas.

When conditions of an emergency character do not prevail, when population density is appreciable, when considerable resources are available, either immediately or in lesser degree over a term of years, measures of a high degree of permanence merit primary consideration. The elimination of breeding areas by drainage or filling, particularly if drainage can be improved by lining or subsoil installation, merits maximum utilization where a satisfactory point of discharge and the requisite gradients are available. Even where drainage conditions cannot be met naturally, mechanical aids, such as pumping from a sump, may permit their application. The fact that a comprehensive drainage program cannot be executed in a short period of time for financial or other reasons, should be no practical obstacle to adoption and initiation of such a program if local administration is sufficiently farsighted to see the advantages of maintaining consistent policies despite changes in personnel. Such comprehensive programs can be executed a single unit at a time in accordance with the general plan and over a term of years. In this connection it is important to plan urban drainage systems, at least in their main channels, with the thought in mind that in future years they may be transformed into storm sewers.

In view of the poor economic conditions of most rural communities, it is unlikely that most rural communities would support an extensive drainage program for malaria control alone, although some opportunities for drainage may exist if reclamation of land for agricultural purposes becomes a possibility. Under these circumstances, therefore, opportunities for permanent sanitation definitely decrease and possibilities of other measures of control must be explored.

In many rural areas, particularly in the tropics, the prevalent types

of houses do not loan themselves to mosquito-proofing. When this is coupled with the intense poverty so frequently encountered, the outlook for the application of more proven permanent measures of sanitation or hygiene, at the cost of the population to be benefitted, is discouraging. Pending their final evaluation and demonstration of practicality, it is under such circumstances that the possibilities of other measures, such as painting houses with DDT and use of aerosol spray, fully merit exploration and study. Naturalistic measures, at times, also deserve consideration. Their character is so varied, and the possibilities differ so greatly with different species of anophelines that their indications cannot be generically discussed.

Mass treatment of the infected persons in a population, either for acute malaria illness or for chronic infections with the idea in view of decreasing the reservoir of parasites, or the administration of drugs to healthy persons for suppression of clinical symptoms of the disease under previous and normal conditions have been procedures, consideration of which indicates a pessimistic implication regarding the applicability of other and more permanent measures. However, future experience with new drugs may alter this pessimistic outlook. Experiences of the war have already taught us, at times, under emergency conditions wide-spread medication is most indicated when it is necessary to suppress clinical symptoms in order to have a force on its feet and functioning.

It may be borne in mind that there is no drug available at present which is a true causal prophylactic, and that by suppressive treatment one cannot be sure of preventing the spreading of infection, although clinical activity most probably will be suppressed for a period of time during which the drug is being taken. By the administration of appropriate drugs armies can be protected for a certain time, against wide-spread clinical manifestations of disease. Under emergency conditions, especially where a large body of susceptibles, with no immunity, must temporarily be introduced into endemic areas, clinical suppression of symptoms will become essential to the maintenance of an effective working force.

The facts elicited by the survey having been balanced against the considerations discussed in the previous section, a choice may be made of the methods most appropriately applicable to the local situation. With this decision made, detailed plans should be formulated for their execution, which may entail further survey work when drainage is indicated. If drainage or fill be chosen as the control measure, the engineer must, as outlined above, ascertain the locations, levels, and profiles for the necessary drainage, the most practicable means to effect the required excavation, and, if ditches are to be lined or subsoil tile installed, calculate the quantities required and their cost, either by purchase or local manufacture. From an estimate of the funds likely to be immediately available, with rough estimates of the probable cost of the entire project, it can then be ascertained whether the whole project can be executed at once or must be spread over a term of years. In some localities it may be necessary to present the project estimated at this point to the appropriating body, in order that the

work can be authorized and the funds allocated. When installation begins, careful records of the cost in materials and labor must be kept in order that unit costs may be computed to aid in the preliminary estimates on further work.

If immediate anopheline control, pending the completion of the drainage program, is required, it may be expected that the entomologist or a competent assistant is concurrently making plans for the supervised distribution of larvicides. His estimates must take into consideration the period of years over which their use will be necessary, the number and area of the localities where they must be applied, the frequency of application, the amounts of different materials required for this schedule, personnel, including both labor and inspectors, and transportation. In addition to the formulation of plans for the systematic distribution of these materials, he must arrange for checks on the efficiency and economical performance of the work. Such checking consists not only in the searching for evidence of anopheline breeding in treated areas, but the search for and capture of anopheline mosquitoes resting in neighboring shelters. Suitable places, strategically related to producing areas, should be sought and designated as catching stations. These are to be routinely visited at frequent intervals. If the catches are zero or low, and the distribution of catching stations has been sufficiently comprehensive, and captures adequately supervised, the destruction of the anophelines in their adjacent breeding places may be inferred. If these are continuously high or the catch suddenly rises from low levels, the necessity for continued or temporary application of larvicide may be deduced, or perhaps a more exhaustive search for new and unfound breeding areas may be indicated. Also in the interim, while permanent measures are being effected, the malariologist must determine other accessory measures which should be used, as indicated by the survey, such measures as treatment of individuals found with parasites, utilization of spray killing solution in places of congregation or where parasite positive cases live and the use of residual DDT inside of buildings.

It is desirable to gather concurrently, where possible during the process of this work, all objective data regarding malaria incidence. Routine spot maps showing the place of residence of all verified malaria patients reported should be maintained. The malariometric survey should be repeated at least annually. It is the basic frame work around which the control program is constructed.

## PARASITOLOGY

### Its Immediate Importance in Control

The parasitological aspect of malaria investigation is of importance as applied to emergency situations common to combat areas, largely with relation to the species, the density, and stage of the cycle of parasite which is present. As previously stated, the presence of P. falciparum is dangerous because of its capacity for rapid clinical action and, when unrecognized, its lethal effect on the host. It has been clearly demonstrated that early recognition of infection with P. falciparum, and immediate and thorough treatment, has resulted in a low mortality among the personnel of the military forces. As long as early recognition is effected, every prospect exists of a continuation of this low mortality in cases of P. falciparum.

Furthermore, knowledge of the species of parasite is of importance to foretell the probability of relapse rates. It has been discovered from the history of the cases of malaria occurring in battle areas that P. vivax is prone to relapse within the wide limits of thirty to eighty percent of all cases, and evidence is available to support the belief that this situation will maintain irrespective of present procedures in administration of suppressive atabrine. With P. falciparum, on the other hand, suppressive treatment, 0.1 gram of atabrine daily, continued over a period of four weeks after removal from malaria transmission areas, frequently results in permanent sterilization of the parasite from the infected individual.

Of equal importance parasitologically is the knowledge of the existence of gametocyte carriers in the area and their location, especially as this is related to areas of the local population frequented by troops and also with relation to the residence of laborers who may be employed in and around the camp area. Furthermore, if residence of persons harboring gametocytes is known, special collections of adult anophelines is indicated at such residences to assist in clarification of the actual vectors of the areas, if these vectors be unknown. These residences or abodes would be the location where the greatest likelihood would exist of finding infected vectors.

Also the presence of high percentages of persons harboring parasites in the sample population at the time of examination and finding of persons with high densities of parasites per field examined indicate either that the disease actually is or recently has been in a stage of active transmission in the locality and that the potentiality for such active transmission exists. Such information is of prime importance to any military installation.

It should never be overlooked that an original diagnosis of P. vivax in an individual does not necessarily preclude the possibility of a later development of an infection where P. falciparum may appear as the principal

infecting agent. A mixed infection may have existed at the moment of original examination in which P. falciparum may have been difficult of easy recognition. Any marked disturbance of mental function on the part of patients suffering from a supposed P. vivax infection should be immediately taken as presumptive evidence of the possibility of an invasion of the parasitological picture by P. falciparum.

## 1. LIFE CYCLE OF PLASMODIA

A thorough understanding of the life of the Plasmodium in man and mosquito is fundamental, not only in the diagnosis and treatment of malaria, but for complete comprehension of established methods of malaria control. Man remains the sole reservoir or "seed bed" of infection despite a continuing search for an animal reservoir, such, for instance, as has been demonstrated to exist with (Jungle) Yellow Fever. The female anopheline mosquito is the transmitting agent, and new species of anophelines capable of transmission are continually being discovered as research in Entomology progresses.

The three species of the Plasmodia described by the Italian workers at the turn of the century are accepted by all authorities. They are: P. vivax, (Benign Tertian Malaria); P. malariae, (Quartan Malaria); and P. falciparum, (Aestivo-autumnal Malaria, Malignant Tertian, Subtertian). A fourth species, P. ovale, has been described, and is now accepted as a separate and distinct entity.

The life of the malarial parasite may be divided into two distinct cycles, (1) the asexual or schizogonous, in man, and (2) the sexual or sporogonous, in female anopheline mosquitoes of certain species. The cycles are essentially the same for all species of parasites.

### a. ASEXUAL CYCLE (MAN)

When an infected mosquito (anopheline female) bites a man, the malaria parasite is introduced into the body in the stage of its development which is known as the SPOROZOITE. Development of the parasitic cycle in man then begins in the reticulo-endothelial system of the body. This phase of life and development inside the human is not completely understood at present. The parasite cannot, in the early stages, be demonstrated in the circulating blood. Eventually, however, the parasites gain entrance into the red blood cells and can be demonstrated in the superficial circulation. When inoculation is made by means of infected mosquitoes, parasites of P. falciparum have been found in the peripheral blood stream as early as six days after inoculation, those of P. vivax as early as the eighth day, and for P. malariae rarely before the twenty-first day. However, the average time of appearance is somewhat more delayed; usually from the tenth to fourteenth day with P. vivax and P. falciparum in natural infections.

The TROPHOZOITE stage, which develops from the sporozoite, marks the beginning of the asexual cycle. The youngest form of this stage is common-

ly called the "signet ring" form, the cytoplasm being arranged in a circular fashion with a small dot of chromatin on the periphery. Actually the parasite is ovoid or spherical, the cytoplasm surrounding a central vacuole, and appearing as circular in optical section. The parasite now begins to perform amoeboid movements, ingesting the available food in the red blood cell. Its growth is gradual, and the shape that it assumes during this period varies with the species. As the trophozoite approaches maturity, it prepares for asexual reproduction, ceasing its amoeboid activity, and assuming a more compact form.

The parasite now enters into the asexual reproductive stage and is termed a SCHIZONT. The chromatin mass of the trophozoite divides into two parts, and progressive segmentation continues until six to twenty-four separate segments have been produced, the actual number of them depending upon the species of the parasite. MATURE SCHIZONT is the term applied to the fully developed and completely segmented parasite, while the immature forms are usually referred to as SCHIZONTS, PRESEGMENTING SCHIZONTS, or SEGMENTERS. The cytoplasm also divides and arranges itself about the chromatin masses. The divisions or bodies thus formed inside the red blood cell are termed MEROZOITES.

When division has been completed, the red blood cell ruptures as the mature schizont bursts, and the individual merozoites are released into the blood stream. Not all of the merozoites survive, but those surviving enter uninfected red blood cells. They, in turn, become trophozoites, thus starting the asexual cycle anew, and the number of parasitized cells in the body increases by geometric progression.

While the majority of merozoites become trophozoites, and continue the asexual cycle, some develop into sexual forms called GAMETOCYTES. They are of two kinds, female or MACROGAMETOCYTE, and male or MICROGAMETOCYTE. Their development is said to take place chiefly in the blood vessels of the spleen and bone marrow, and immature forms are only occasionally found in the circulating blood. Gametocytes grow more slowly than the asexual forms, requiring about twice as long to reach maturity. They are capable of reproduction within the body of the female anopheline mosquito, but not in the human body. If they are not sucked up by a mosquito while taking blood, they die, it is thought, within a few days.

The asexual cycle, from the youngest trophozoite to the release of the merozoites, is completed in forty-eight hours by P. vivax, P. falciparum, and P. ovale. That of P. malariae requires seventy-two hours.

#### b. SEXUAL CYCLE (FEMALE ANOPHELINE MOSQUITO)

In order that the mosquito become infected, the person with malaria must have a sufficient number of gametocytes in the blood, and it is said male and female forms must be ingested in proper proportions by the mosquito. After the parasites are taken into the mosquito's stomach, while taking her blood meal, all forms of the parasite, except the mature gametocytes, are

digested. The latter continue their development, the microgametocyte (male) undergoing exflagellation, and the macrogametocyte (female) casting off chromatin polar bodies. The resultant products, microgamete and macrogamete, respectively, now make contact, fertilization takes place, and the ZYGOTE is formed. This body elongates, becomes motile, and is termed an OOKINETE. It now migrates through the stomach wall, coming to rest between the outer membrane, and the inner epithelial layer, and then contracting to a small round body called an OOCYST. The oocysts may be readily seen on the stomach wall by microscopic examination. The oocyst grows tremendously, the nucleus multiplying by repeated divisions. The cytoplasm arranges itself about the nuclear divisions and SPOROZOITES are formed. When mature, the oocyst ruptures, releasing the many sporozoites into the body cavity of the mosquito. They are long and thin, with a centrally placed nucleus, and they spread throughout the body of the mosquito, many finally resting in the salivary glands. Consequently, they are injected with the saliva when the mosquito bites the human subject, thus initiating the asexual cycle in man. This entire process of the sexual cycle in the anopheline mosquito requires approximately 10 days to 3 weeks, depending on the species of mosquito involved, the species of parasite, and various meteorological factors.

Concepts of  
Particular Phases of Control

## DDT AND ITS USES

### FOREWORD

Practically all discoveries of merit, particularly in the medical world, are heralded as the panacea, the "cure-all" of existent undesirable conditions. The more the actual merit of the newly discovered agent or procedure the greater are the claims of infallability for producing desired results. There have been in the past so many examples of discoveries substantiating the above statement that individual justification of the statement need not be advanced.

The discovery of the lethal capacity of DDT against certain insects has offered no exception to this rule. Human nature ever searches for Utopia. In the mind's desire, the days of miracles have not passed.

DDT is an excellent addition to the arsenal of agents directed against insect borne diseases. However, scientific evaluation of its capabilities in proven phases of disease prevention should replace over enthusiastic hopes and expectations as the criterion of results accomplished.

## DDT AND ITS USES

### 1. HISTORICAL

DDT was first synthesized by a German chemist, 1874. It is also known under the names "Gesarol" and "Neocide". Its abbreviated name "DDT" is derived from the chemical formula which, stated simply, is "Di-chloro, Di-phenyl, Trichlorethane" or more accurately "2-2 Bis (P-Chlor-phenyl) 1-1-1 Trichlorethane. Insecticidal properties were not discovered until 1939 when such properties were demonstrated by a Swiss Chemical Firm, Geigy and Company. The United States Department of Agriculture tested a sample in 1942 and found it effective against many insects.

### 2. PROPERTIES

It is a fine, white powder, having a tendency to lump. As supplied ordinarily, commercially, it is not in pure form. It is insoluble in water, but is soluble in:

Solvent	Grams DDT per 100 C.C. Solvent
Cyclohexanone	100-120
Benzene*	77- 83
Trichlorethylene*	72- 83
o-Dichlorobenzene	63- 71
Tetrahydronaphthalene	63- 71
Ethylene dichloride*	56- 62
Xylene*	56- 62
Acetylene tetrachloride**	55- 62
Acetone	50- 55
Polymethyl naphthalenes	45- 50
Carbon tetrachloride*-***	46- 48
Benzyl benzoate	39- 41
Fuel oil No. 2	Approximately 10
Cottonseed oil	" 9
Stoddard solvent	" 9
Kerosene, crude	" 8
Fuel oil No. 1	" 8
Kerosene, refined, odorless (Deobase)	" 4
Ethyl alcohol	" 1.5

\* - Air Corps Issue

\*\* - Chemical Warfare Service Issue

\*\*\* - Quartermaster or Corps of Engineers Issue

### 3. TOXICITY TO INSECTS (GENERAL)

DDT has a powerful toxic effect on many insects through its action on their central nervous systems after its absorption through the chemotactic

sensory organs in the tips of their tarsi in adult stage as well as by direct contact in larval stage. DDT does not repel. All mosquitoes tested and probably the majority of insects will readily rest on it. The insects which suffer mortality on sufficient contact with it include several species of diptera, human lice, cockroaches and grasshoppers, Japanese, as well as other destructive beetles and weevils, goat lice, some caterpillars and moths, termites, bedbugs and squash bugs, plant aphids, thrips, dog fleas, and sand fleas. All facts regarding lethal qualities against insects of importance to the military have not, as yet, been thoroughly studied and proven, but certain truths can now be accepted. DDT manifests its lethal capacity not only at the actual time of application, but also retains this same capacity over long periods following application. This latter manifestation in which lethal qualities are retained for long periods, killing insects which happen to make contact with it long after the time of its original application, is known as the residual action. Against adult albimanus and aegypti, its killing capacity is greatly augmented when the DDT remains in solution. (See later in the memorandum discussion of experiments being conducted by the Army School of Malariology).

As far as is known, DDT as a larvicide will kill all types of mosquito larvae with which it makes contact in adequate quantities. Its action on larvae is believed to be by actual contact with the crystallized or dry DDT particles as well as while in solution in various solvents. It does not kill "in solution" in water as it is insoluble in water.

As an aerosol and air spray, it also will definitely kill mosquito adults with which it comes in contact in sufficient quantities for sufficient time.

As a solution or an emulsion painted on beds, springs, or mattresses, it will kill bedbugs.

DDT, used as a dry powder or impregnated in solution into clothing, will kill body lice. Two percent emulsion of DDT is most commonly used for impregnation. Underwear so impregnated contains approximately fifteen grams of DDT per two-piece-suit or two percent of the weight of the garment. These garments customarily remain effective even after as many as six washings. A DDT emulsion for use as "suds" can be made as follows:

To 1 gallon of concentration of DDT, which is 25% DDT, 68% xylene (solvent) and 7% triton x-100 (emulsifier), add 11 gallons of water. The 12 gallons resulting will suffice for approximately 35 two-piece underwear suits (50% wool).

The importance of DDT in a military program can be well realized when it is recalled that Malaria, Yellow Fever, Dengue, Filariasis are all carried by mosquitoes, and Typhus is carried by the body louse. These diseases constitute some of the greatest hazards from the standpoint of a war effort.

#### 4. TOXICITY FOR MAN

Since DDT is more readily absorbed when in solution than when in dry form, solutions in oil and organic solvents are more dangerous than the dry powder from the standpoint of toxicity through absorption. Continued application of DDT in solution to the skin of animals, as well as when added to the diet, has resulted in toxic symptoms and definite damage to organs. The symptoms comprise:

Anorexia  
Loss of weight  
Hyper-excitability  
Tremor and convulsions

The pathological damage to organs and tissue include:

Degeneration of anterior root neurons and  
toxic necrosis of liver and kidneys.

However, in practical field use there are practically no records of any severe symptoms developing in personnel because of such use. In animals 100 mgs. of DDT per kilo of body weight, when taken by mouth, produced no toxic effects. Two hundred (200) mgs. per kilo per day produced death in 90% of mortality in test animals. (Time over which this dosage was taken was not stated). This dosage would ordinarily be greatly in excess of any amount man would acquire in practical ordinary use of the substance in the field. However, it is wise to observe certain precautions:

- a. Inhalation of dusts, sprays, or mists should be reduced to a minimum.
- b. Foods should be protected against exposure to and contamination with DDT when sprays are being used. Also, the place of storage should be carefully chosen as DDT resembles certain powders used in cooking, is tasteless, and accordingly, it should never be stored with food materials or in kitchens.
- c. Certain solvents are particularly dangerous, as systemic poisoning can result from inhalation of the solvent itself. Also, dermatitis may develop. These special and dangerous solvents are:

tetra chlorethylene  
carbon tetrachloride  
benzene

acetylene tetrachloride  
ethylene dichloride  
trichlorethylene

#### 5. DDT AGAINST MOSQUITO ADULTS

DDT is being used against adults in three generally different manners:

- a. As a spray (an aerosol) in individual hand sprayers used inside of

dwelling and in small scale outside of buildings. (In this type pyrethrins are usually combined with DDT for quick knock-down).

- b. As an airplane spray for large areas in the form of aerosol and also as air spray to produce residual deposit.
- c. Painted or sprayed on areas, walls, screens, inside of buildings, and also sprayed outside on vegetation where mosquitoes will subsequently rest and come in contact with it and absorb the poison.

Mention should now be made relative to the distinction between an aerosol and a spray. There should be a clear understanding of the difference between an aerosol and an air spray. Technically the difference is determined by the size of the droplet involved. Speaking in a general manner, droplets in size from 1 micron to 50 micra would be considered in the category of belonging to aerosols. Whereas, when sprays are composed of droplets from 50 micra and up to several hundred micra, the resulting droplet mist would be considered to be a spray.

Practically, of course, there is a distinct difference in the manner of functioning of aerosols and air sprays. The active ingredients of the aerosol float in the air for many minutes and sometimes even hours after application during which period flying insects naturally have an opportunity to fly into it and make contact with it. An air spray, on the other hand, falls relatively rapidly to the ground when delivered from an apparatus applied by hand or by airplane. The lethal potentiality of the active ingredient will then depend largely upon the amount of coverage of vegetation which is secured and the subsequent contact insects make with it. When considering a forested area, the amount of foliage at levels high above the ground, which can intercept a spray applied by airplane, can readily be appreciated. Subsequent kill of insects would depend not only on the elevation above the ground where these insects rest and live, but also by the actual contact made with particles of DDT either crystallized out of solution or still remaining in solution.

An aerosol, on the other hand, applied in any manner, by ground sprayer or by airplane, would remain suspended in the air for considerable time where flying insects could make contact with it. Moreover, under these circumstances, greater opportunity would be afforded for penetration of the jungle foliage by the aerosol to differing levels, dependent, of course, on air currents and on winds.

#### 6. THE AEROSOL FOR INDIVIDUAL OR PERSONAL USE

The bionomics of the mosquito in question must be known and understood before spray killing can be utilized to its highest degree of efficiency. Those mosquitoes which rest inside of habitation while searching for, and after securing a blood meal, would be particularly susceptible to use of aerosols inside of houses. Pyrethrins are practically always included in such aerosol sprays for the quick "knock-down" and DDT is added for the

purpose of its residual effect. Oil of Sesame is included, together with pyrethrum, as an activator, and the freon, naturally, is simply the vehicle and source of expressive power, the expellent. When used in combination, a common formula is:

3.0% DDT  
0.4% pyrethrins  
5.0% cyclohexanone  
5.0% #10 lubrication oil in Freon 12

## 7. OUTDOOR SPRAYS OR AEROSOLS FOR ADULT MOSQUITO KILLING

This subject is still under investigation. If DDT actually comes in contact in sufficient quantity with adult mosquitoes, as stated above, it will kill them. With air spray from airplanes, it is difficult to be sure of the "coverage" secured as well as the universal dosage deposited on ground vegetation.

Also, the droplet size, which differentiates between sprays and aerosols, appears to be of importance in this killing effect. Is the aerosol droplet size, 1-50 micra, the most effective, or is the spray size, 51 micra and up, the better, or is the best type of application a combination of both sizes? Investigators have stated, as based on experimental data, that with 8% DDT in oil, the most effective micron size is that with a radius of 5 micra.

It is still a debatable question whether spraying from airplane in the customary manner used at present can effect an appreciable kill from the action of residual DDT which is deposited on the vegetation after one spraying at the rate of 0.6 lbs. per acre. Here again, it is the question of the amount of coverage acquired on the vegetation during the spraying, and the subsequent period of contact made by mosquitoes with this residual deposit.

For practical use, the question is whether sprays kill by their action as aerosols or by residual action when on the vegetation, and also how to apply the spray to realize the most efficient droplet size to produce sufficient quantities for sufficient time to realize the greatest lethal effect. The subject of the best solution to be used is still partially in the experimental stage. The status of the most efficient size of the droplet is still under investigation. The demonstration of the actual percentage of unequivocally proven kill is still being investigated.

In view of the fact that investigations are still in progress to determine the best manner of application of DDT to secure the highest efficiency of kill, it is appropriate to mention the varying means of measurement employed to determine such efficiency. The most important measuring rod is naturally the lethal effect of the solution on mosquito larvae and adults, particularly anophelines. This measurement can be realized in a number of ways. For use of future investigators, a description

of detail of procedures of measurement of results appears indicated.

a. Experimentally planned determination of mortality.

(1) Larvae

(a) Larvae can be placed in suitable containers in the area to be sprayed in a manner to cover an adequate sample of the area and also to represent differing types of coverage (foliage). Two containers should be placed at each station selected, one pan with larvae left uncovered, and a pan of same size and same number of larvae, placed close to the test pan, but carefully covered so that no DDT can enter. Observations of the test pans over a 48 hour interval, after application of DDT, will show deaths of larvae, which, when compared to the covered comparison pan will indicate whether larvae deaths were due to the spray solution or to natural causes. (Note: The covers should not be removed from the comparison pans except by a trained observer with the greatest precaution. Also, no sticks lying in the vicinity of the pans should be used to stir up larvae to ascertain if dead or alive. Both of these procedures could introduce DDT into the comparison or test pans from a chance particle of DDT having fallen on the object so used. Furthermore, a rod, if made of glass, when used for stirring one pan of larvae, should be carefully washed and dried before using in another pan, in order to avoid possibility of carrying particles of DDT from pan to pan. Probably a better procedure is utilization of wooden applicator sticks for stirring, and breaking and discarding the stick after use at each pan.

(b) A better procedure to be followed with larvae, in order to avoid other causes for death of larvae in the field, such as effects of transportation, of handling, of meteorological influences, is to expose plates or pans with no larvae in them, but with water added, or perhaps even without water, at stations selected comparably to those noted above. The pans should be carefully identified by suitable numbers. If water is used, this is poured, after a suitable interval following the spraying, into a labelled bottle. If no water is used, the pan is protected and brought to the laboratory.

If water has been used, it is poured from the bottles again into the original pans in the laboratory, and if dry pans were used, water is added to them. Then larvae are introduced into all pans, and subsequent deaths can indicate the presence of DDT.

The larvae under this latter situation will not have been exposed, with resulting equivocal interpretations, to other factors in the field producing mortality, such as those mentioned above of transportation, handling, and meteorological influences.

(2) Adults

These can be exposed in cages at strategically selected points and an

additional cage with comparable mosquitoes protected against the entrance of DDT solution placed near the test cages. These latter will serve as comparison cages. Comparison cages could also be placed, unprotected by covering, in areas where meteorological factors are comparable to the area to be treated, but where spraying is not to be performed. In this manner either set of comparison cages would serve to indicate if the DDT solution were the cause of any mortality in the treated cages.

- b. Observations in nature to determine lethal effect of DDT solution on larvae and adults.

Perhaps the most conclusive method of demonstrating killing effect is the record of the mortality as it actually occurs in the larval and adult fauna as they exist in nature.

The final determination of such results is measured by the presence and density of larvae and adults before and after application of DDT solution. Determination of the status of larval presence and density prior to treatment is more or less objective as larvae by their nature are limited to water areas and limited in transporting themselves long distances. A good observer can check larvae density prior to and post to any procedure with good prospect that his observation will represent a fairly accurate sample as it exists in nature.

The sampling of an adult population presents greater difficulties as locomotion over large areas on wing is relatively unlimited, and presence of adults in any one locality at a specific time is dependent upon variable factors, which may invalidate the findings as these are interpreted to indicate actual numerical presence of adults in nature. Such influencing variable factors include:

- (1) Distance of station from breeding spot.
- (2) Type of place selected for capture station and its degree of:

Dampness.  
Protection from winds.  
Shade.  
Source of blood meal.

- (3) Termination of or beginning of breeding season.
- (4) Density of breeding in breeding area.
- (5) Wind direction and velocity.
- (6) Humidity and atmospheric pressure.
- (7) Sunshine, moonlight, shade.
- (8) Rains.
- (9) Temperature (heat and cold waves).

Alteration of any of these factors may drastically change results

of captures from those previously performed even when no outside procedure to cause mortality has been applied.

Negative findings in a station at any one specific time, or in consecutive captures, may not represent a true sample from the general region, but merely represent a sample from a particular capture spot influenced by a change in some of the above mentioned factors.

For these reasons, when attempting to evaluate relative decrease or increase in mosquito population, it is essential to establish in nature a comparison area where factors influencing the presence of mosquitoes are comparable. Captures are made under the same conditions then as in the area to be treated and observations are made over a period sufficient to judge the normal density of mosquitoes in the area and its variations as these occur ordinarily in nature. When the captures in the comparison area show wide variations under normal conditions, results attained in treated areas, after treatment, have to be viewed with caution in the light of these normal findings in the comparison area.

If, and when the sampling procedure indicates a degree of stability in both comparison area and areas to be treated, then conclusions can be drawn with expectancy that they represent the true picture of actual mosquito prevalence in the area.

However, facts, at present at hand, warrant, under indicated circumstances, the trial of spray killing of adults by airplane spraying or other types of spraying apparatus.

TB Med 110, October 25, 1944, page 4, paragraph 5, states:

"Undue optimism as to the value and properties of DDT as a cure-all of mosquito control problems is to be discouraged. DDT is not a panacea. Unrelenting observance of all aspects of malaria control is mandatory to which we may expect DDT to add significant improvement in results."

Quoting again, page 4, paragraph 6:

"In those areas where a heavily infested native reservoir must be dealt with and where vector species prefer indoor resting places, the treatment of native dwellings should take preference over other uses followed in order of priority by the treatment of U. S. Army Forces domestic installation (barracks, dayrooms, messhalls, tents, indoor latrines, etc.) and the larviciding and spraying of vegetation in and around camp sites. In those areas where a native reservoir does not present an immediate threat or where a species preferring out-of-door resting places is encountered, the treatment of U. S. Army Forces domestic installation and employment of DDT as a mosquito adulticide out-of-doors should be given first consideration."

Today the most commonly used solution is 5% DDT in oil, usually fuel oil #2 or kerosene. (See TB Med 14, 3 March 1944)

From the results of experimental work performed by the Army School of Malariology, it would appear, as previously noted, that the time element of necessary contact to produce a kill with DDT is greatly increased when the agent crystallizes out of solution or is naturally in a dry state. It would appear that DDT crystallizes out of kerosene much more quickly than when in solution in Diesel oil #2. For this reason, it is possible that Diesel oil #2 is a better solvent than kerosene for ordinary usage. In theaters of operations it should be about equally available. Other solvents will hold DDT in solution for longer periods than kerosene or Diesel oil. They are mineral oil, SAE #30 and SAE #50. They are not, however, as readily available.

#### 8. KILLING BY INSECTS ALIGHTING AND RESTING ON DDT COATED SURFACES

It has been clearly demonstrated that mosquitoes, (A. aegypti, A. taeniorhynchus, A. albimanus), lighting for sufficient time on surfaces which have been painted with a DDT solution in sufficient quantity and correct manner will die from the effect of DDT absorption. From tests conducted at the Army School of Malariology, it is clear that the potency of this killing effect is dependent on certain factors. Different genera and species of mosquitoes are effected in varying manners where the same period of contact is used and mortality is recorded over the same period following time of contact.

The solvents which are utilized are an important factor. Accordingly, a long lasting, slowly evaporating solvent is to be desired. In order of their capacity for holding DDT in solution and, accordingly, producing a quicker kill with lesser periods of contact, the solvents tested by the Army School of Malariology are listed below in order of their efficiency, the best being stated first.

SAE #30 and #50  
Mineral oil  
Diesel oil #2  
Kerosene  
Acetone

Acetone is a very poor solvent considered from the standpoint of its capacity for holding DDT in its dissolved form. Kerosene is somewhat better, whereas SAE #30 and #50 and mineral oil are all comparable and decidedly better than kerosene or Diesel oil #2.

A series of experiments, which is still in progress, was planned by the Army School of Malariology to demonstrate periods of resting contact with varying dosages of DDT in varying solvents.

Screen wire was painted with DDT in solution in dosages including

50, 100, and 200 milligrams per square foot. The solvents selected for trial were acetone, kerosene, fuel oil #2, Mineral oil, SAE oil #30 and SAE oil #50.

The pieces of screen wire painted with varying solutions were tested at intervals after painting by adjusting the screen around a bare arm and applying colony-reared A. aegypti and albimanus and wild captured A. taeniorhynchus and A. albimanus. Each mosquito, in a test tube, after a period of no blood meals, was offered a blood meal by inverting these test tubes, and applying the open mouth to the screen wire. When alighting to bite, they alight on the screens, variously treated with DDT.

From the deaths resulting after such exposure to DDT in varying solvents at various periods after painting, the following facts were observed:

After four instantaneous contacts\* with DDT in solution, a very high percentage of mosquitoes will die with characteristic signs of poisoning within 24 hours after such contacts.

From the limited number of tests so far performed, the length of time the various solvents retain DDT in solution under the conditions of the experiment (Panama wet season meteorology with screens kept inside open screened buildings) and accordingly effect a high percentage of kill on four contacts is as follows:

Acetone not over 36 hours.  
Kerosene not over 72 hours.  
Diesel oil #2 up to 39 days.  
Mineral oil (one test only) up to 73 days.

It would appear that meteorological influences may have a bearing on the length of time over which certain solvents can hold DDT in solution. Moisture in the air appears to favor retention in solution for a longer period, and less moisture, with accompanying dry season and winds, appears to favor an earlier crystallization.

The optimum dosage for such painting for residual effects has not, as yet, been fully determined, but the range of dosage would appear to be somewhere between 25 and 200 milligrams per square foot. In the experiments so far concluded at the Army School of Malariology the 200 milligrams per square foot dosage has been found superior in its lethal capacity to the 100 milligrams and 50 milligrams dosage.

\* Instantaneous contacts can be realized by slipping a card between the mouth of the test tube and the wire at the instant the mosquito alights upon the wire to secure a blood meal.

DDT solutions ready for military use are listed in WD Circular #151, 1944.

Stock No.	Item	Unit
51-1-305	Insecticide Spray, DDT Residual effect	Gallons
51-1-120	Larvicide DDT, powder, Dissolving	Pounds
51-1-122	Larvicide DDT, powder, Dusting	Pounds

The first listed is all ready for use as a spray.

The second listed product can be made into a solution of 5% in kerosene to prepare a product identical with the first. Crude kerosene is better than the highly refined product.

The manner of application of solutions should vary, as mentioned earlier, in accordance with habits of vectors. Bionomics of species of anophelines must be understood. A species with sylvan habits will not be killed in any great percentages by painting with DDT inside of sleeping quarters or buildings in general. For those anophelines which rest inside of buildings, spraying or painting the inside of buildings with the solution would have a beneficial effect in killing mosquitoes. For the sylvan variety, better results would probably be obtained by spraying the vegetation in and around the camp by means of air sprays (power or hand) or by airplane. Knapsack and flit guns, 2 quart variety, sprayers can be used. At times, to certain surfaces, the solution can best be applied by a paint brush, such, for instance, as those used when painting screen wire.

For residual effect the spray should be relatively coarse. Too fine a spray floats; too coarse a spray falls immediately to the floor.

After treating surfaces with DDT, tests should be made from time to time with caged mosquitoes placed against these surfaces to determine over what period of time killing properties are still existent. One side of the cage can be made removable so that mosquitoes may rest upon the wall painted with DDT, after the cage is placed in contact with it.

As a spray for residual effect, it is recommended, for the present, until better information is available, to apply at the rate of 200 milligrams per square foot, or 1 quart of a 5% spray solution per 250 square feet of surface. It is believed, under normal conditions, that effective killing will be maintained for approximately three months after one such application. (Note: A field experiment is now being conducted by the Army School of Malariology to determine accurately the period over which such

treatment of surfaces with DDT will retain their lethal effect.

Although DDT powder, as noted above, does not act as quickly in its residual killing capacity as when in solution in a spray, it should be used when solutions are not available. Ten percent stock DDT powder should be diluted with suitable dust diluents to produce a 2% solution (1 part powder and 4 parts diluent dust). Approximately 3 pounds of the 2% mixture will be required for 1000 square feet, a dosage of 30 milligrams of DDT per square foot.

Where surfaces have been painted or white-washed, the residual killing action of DDT may be reduced. This situation should always be carefully noted. In the case of dusting with the 2% mixture, the most suitable item of military equipment is the duster insect powder (Q.M. stock #41-d-3750). When dusting in the interior for residual effect, it is recommended that the surface be first treated with water. The duster is then held approximately one foot from the surface and directed upward or downward at a 25° angle.

#### 9. USE OF THE INSECTICIDE AS A MOSQUITO ADULTICIDE OUT-OF-DOORS

With studies still in progress at the present time, it is impossible to be dogmatic relative to this subject. However, the results so far demonstrated would warrant the use of such spraying under local circumstances, such as to protect bivouacs, gun replacements, observation posts, out-of-door moving picture theaters, and other points of assembly. Present knowledge, furthermore, merits attempts for the reduction of the adult mosquito fauna of larger areas through the medium of airplane spraying.

It is claimed that a reasonable reduction of adults can be secured through the use of 0.1 pound of DDT per acre in a 5% kerosene or Diesel oil #2 solution. (See TB Med 110, 25 October 1944). This would equal 1 quart of the 5% solution in oil. However, residual effects are not anticipated with such a low dosage. In all probability, it would necessitate 2 or 4 pounds of DDT per acre (about 5 to 10 gallons of 5% solution) to realize appreciable residual effects.

It should be remembered that weather effects, such as rains, high winds with dust, will have a detrimental effect on the lasting residual DDT action. As mentioned above, where airplanes are not available, other types of sprayers with power equipment may be utilized. Frequently, these may be transported by truck.

#### 10. DDT IN OIL AS A LARVICIDE

In its use as a larvicide, it would appear that the information so far at hand relative to evaluation of results and dosage is more exact than that pertaining to its use as an adulticide. This situation in all probability results from the fact, as before mentioned, that measurement

of results by larval observations is a more exact procedure than measurement by sampling adult mosquito populations. Once again, it is a demonstrated fact that DDT in sufficient quantities in contact with larvae for sufficient time will absolutely function as a larvicide. Here again, the solution in most common use is a 5% DDT solution in kerosene or Diesel oil #2. Not only is this the most common mixture, but probably the most easily obtainable in practically all theatres of operations. It has been estimated that 1 quart of a 5% DDT solution will treat effectively approximately one acre of water surface; whereas, 10 to 14 gallons of oil without DDT added are considered as necessary to produce effects somewhat comparable. A group of special investigators have recently recommended, though not in published form, the use of 5 quarts of a 1% solution instead of 1 quart of a 5% solution. It is claimed by them that coverage is easier and results more efficient with this preparation. To prepare a 5% (weight/volume) solution commercially pure DDT is dissolved at the rate, roughly, of 7 ounces to 1 gallon of oil. Kerosene, fuel oil, Diesel oil, and even, under emergency conditions, waste crankcase oil are utilized as diluents. The resultant mixture should be stirred at intervals until all DDT is in solution. This may necessitate twenty-four hours. Moderate heating of this oil, considerably below the boiling point, facilitates solution.

In determining actual dosage for specific areas and types of areas, it is advisable to make larval observations and record results to determine the most effective, at the same time, economical dosage.

When used as a larvicide for residual effect, the subsequent kill of larvae will depend upon the period of time over which the DDT remains in the place where larvae are present. This stability of location will depend, in turn, largely on the amount of vegetation that exists and the absence of winds and current which favor its removal. For residual use as a larvicide, it is customary to apply 5 gallons of a 5% solution in oil per acre, which is equivalent to approximately 2 pounds of DDT per acre. (See TB Med 110, 25 October 1944).

Regarding the method of application of DDT oil solution as larvicide, this will depend largely on the equipment at hand. Whatever equipment is used, the spray nozzle should be adjusted to produce a fine spray. Various types of knapsack sprayers can be used, and at times, under certain conditions, even the flit-gun type or the paint sprayer with power equipment will serve. Sometimes the solution can be poured directly from a container, and, where spreading qualities are favorable, good control can be secured. Sand and other comparable material can be saturated with the solution, and thrown over the water surface comparable to the manner of sewing grain. Sand and sawdust, soaked in the solution and placed in burlap bags, can also be moored in the bottom of pools or in streams.

#### 11. DDT DUST AS A LARVICIDE

It is apparently agreed by most observers that DDT as a larvicidal

dust has not the general utility under war time conditions as DDT in oil solution. The preference for the oil solution is due to its greater ease in handling. However, DDT powder is claimed to be considerably more effective in its action than Paris green. "Larvicide DDT Powder Dusting" is supplied by the Quartermaster as a 10% mixture in talc. A 1%-5% dilution is most frequently used. For a 1% mixture, add 1 part of this concentrate to 9 parts of dust diluent. Soap stone, condemned flour, or road dust (dry) can be utilized as effective diluents for the dust. In making dust dilutions care should be taken that all lumps of DDT, resulting from packing and storage, are carefully broken. Application is similar to the application of Paris green, by means of the prescribed dusters, or by throwing on by hand. The dosage is usually 1/10 pound per acre, or 10 pounds of 1% dust dilution. The dust dilution may also be tried for killing of adult mosquitoes out-of-doors. Five percent dust mixture is most frequently used for this procedure, customarily applied from a duster to the vegetation surrounding the area to be protected. Such an area may be a gun emplacement, fox hole, or other locality where men remain for a period of time. It is customary to apply the dust to a zone with radius of 50 to 100 yards around the spot to be protected.

## MAIARIOLOGICAL ASPECTS OF SPRAY KILLING OF MOSQUITOES

Spray solutions for killing of adult mosquitoes have been in rather common use for a decade or more. The active ingredient in these sprays at the present time is principally pyrethrin, an extract of pyrethrum flowers. The chemical, DDT, is at times used in combination with pyrethrin because of its long lasting qualities.

The general value of these sprays has been recognized during the period over which they have been in use, but in recent years evidence of a more scientific nature has been advanced to demonstrate conclusively their value as an additional agent for malaria control, at least when directed against certain species of anophelines.

Russell and Knipe conducted a series of experiments covering three years observation in India, demonstrating the efficiency of pyrethrum in malaria control by spraying once a week under most unfavorable conditions, namely, in native thatched huts. The effectiveness of the program was measured by the spleen and blood index. Their results were very conclusive. The species against which the procedure was directed was A. culicifacies. It was demonstrated during the course of the experiments that spraying twice a week produced more pronounced results than spraying once a week.

In discussing the use of spray solutions as a practical method for the control of malaria, it is perhaps indicated to review theoretically the factors upon which the efficiency of spray-killing depends.

In the first place, effective spray-killing can have an immediate effect on preventing infections with the malaria parasite because these solutions, if used immediately on arrival in houses in infected areas, will kill in a few minutes anophelines already infectious. For this reason, it is indicated that spray-killing apparatus should accompany all expeditions, and should be immediately used, at least on entering all houses or shelters, if routine pre-spraying before occupancy proves impossible.

Special attention should be given to places of congregation, such as restaurants, hotels, bars, moving picture houses, etc., in the local habitated areas where there is expectancy that personnel of the expedition may congregate.

In the second place, spray-killing can be effective in killing uninfected mosquitoes before they have had opportunity of biting infected man, and also in killing mosquitoes which have bitten infected man before sufficient time has elapsed for them to become infectious. In other words, spray-killing can reduce the reservoir of parasites actually present in

the vector of the area, and can prevent new infection developing in new potential vectors, so reducing the general infected vector density of the area in question.

It is theoretically possible even to eliminate the parasite from an area by spray-killing. The cycle of life of the parasite existent in the mosquito can be killed and without new transmission, because of no infected vectors, the parasites in human reservoirs can die as a result of protective bodily reactions. New parasites could, of course, be introduced into the area.

The effectiveness of spray-killing inside of houses depends naturally upon the ingredients which are used, their concentration and condition, the type of spray gun and effectiveness of the personnel using the spray. Success of indoor spray-killing employed as a routine application depends primarily upon the length of time a given species remains inside the house, also how frequently the mosquito, after taking its original blood meal and before it becomes infectious, returns to a house which is being sprayed routinely.

The activities of a newly hatched female adult, in chronological order of events, is generally considered to be a short period of inactivity in the neighborhood of the breeding area for one or two days after emergence, subsequent mating, and then the search for a blood meal, a period of rest while engorged with blood, a search for a water area for oviposition associated with fertilization again, and then back for another blood meal. While the mosquito is inside a shelter, that, of course, is the most opportune time for it to be killed with spray-killing solution. Theoretically the ultimate success of spray-killing in houses would depend upon the length of time, or number of times, the anopheline would be inside houses, so affording opportunity of coming in contact with the spray solution.

It would seem that the success of the spray-killing demonstration conducted by Russell and Knipe of India, with spraying only once a week, would, from a theoretical standpoint, not customarily be reduplicated unless special conditions existed, such as a species largely domesticated, or a rather inefficient vector, or, through the circumstance of working in localities in which houses presented the most suitable resting place. It must be remembered, however, that the results produced in Russell and Knipe's work did not involve an immediate reduction of malaria incidence to a low level. Results were accumulative over one to three years.

One element in the success of the recent elimination of A. gambiae from northern Brazil was undoubtedly the fact that spray-killing solutions were highly effective against a domesticated mosquito. When dealing with anophelines of less domesticated habits, or with host preference preferably zoophilic, spray-killing inside of habitations would not be as successful. Under customary conditions, precaution would indicate that the number

of spraying per week be increased to offer greater opportunity of contacting the mosquito with the spray, and resultant killing of the adult before sufficient time has passed for it to develop infection and be capable of transmitting it. Complete assurance of success in spraying, and with a partially domesticated anopheline, could probably be guaranteed by spraying twice daily, just after dusk, and just before dawn, in habitations, latrines, guard houses, etc. When dealing with species partially, at least, zoophilic, stables and their environs in vicinity of habitations should be sprayed possibly bi-weekly.

Spray-killing, when utilized as a means of malaria control, may appear to represent an attack of the problem at a link of the cyclical chain of malaria transmission previously considered to be illogical, namely, concentration on the elimination of the individual mosquito either already infected or potential for infection, while leaving the constant source of anopheline vectors from breeding areas practically the same. However, its importance should never be under-estimated as it may prove to be one of the most economical and potent tools in the fight against malaria, comparable perhaps to the long sought drug which should be cheap, effective, quick in action and potent against the sporozoite.

Spray-killing solution should also be used against mosquitoes carried in transports, trains, ships, planes, vehicles, coming from infected areas in order to kill possible infected adults which have come on board, and, in order to prevent the introduction of a new species into a new area, as occurred when A. gambiae was introduced into Brazil from Africa by transportation across the ocean. The killing of infected adults, introduced on board the vehicle of transport, will prevent the infection of those on board this vehicle by these anophelines, and also prevent the possible introduction of a new parasite or new anopheline species into a new area.

It is believed by some that pyrethrum has a repellent action, and when sprayed, will tend to deter the entrance of mosquitoes as well as kill those which actually come in contact with it. The status of pyrethrins as repellents, however, is not as yet entirely clear. It has been demonstrated by Elmendorf and Marucci (unpublished) that pyrethrum in high concentrations has, at least, a residual lethal effect for several days in killing mosquitoes introduced into a cage previously sprayed.

For economy and efficiency of operation, special attention should be given to the apparatus used for spraying. The resultant spray should be fine and light so that it will float and there should be no drip from the nozzle. The drip from the nozzle, not only constitutes waste, but also can produce damage to belongings and create dislike for the procedure where such damage would be of concern to the inhabitants. In war situations supply of pyrethrum may be limited and available transportation is always a problem. The solution should not be wasted. The spraying solution should be tested for its lethal effect against the anopheline vector of the region. For such a test, caged anopheline vectors are recommended. These should preferably be

reared in insectaries to insure that they are not infected with malaria parasites. Pyrethrins, if potent, should kill all anophelines exposed to direct contact with it inside of a period of three minutes.

Various types of spraying apparatus are available, among them being hand-pump sprayers with continuous spray and intermittent spray, motor-driven pressure sprays, hand-pump pressure sprayers, can sprayers pressured by a refrigerant gas, and the Westinghouse Pyrethrum-Freon Bomb.

The Westinghouse bomb is most compact, consisting of a cylindrical metal container approximately five inches long by two and three-quarter inches in diameter, weighing about 1 3/4 pounds and equipped with a screw cap which opens and closes the vent through which a fine spray is blown by its own content of freon. The bomb will function for 12 to 13 consecutive minutes. Four seconds of spraying will suffice for 1000 cubic feet of area and three seconds per pup-tent. The containers, at present, are non-refillable and the bomb is expendable. All other factors being equal their use should be confined to work of an emergency nature such as the case of troops on the march, whereas a type of pump sprayer with an efficient atomizer should be utilized in established or semi-established bases.

## MOSQUITO PROOFING

In common parlance, the protection of a building against the entrance of mosquitoes is loosely designated as "screening". This expression is not exact. The protection of a building against the entrance of mosquitoes and flying insects should be defined as "mosquito-proofing". This expression implies that more work is usually involved than the simple expedient of application of screen doors to doorways and screen wire or screens over the windows.

It has been generally accepted that mosquito-proofing will limit infection with malaria. General Carter in 1914 stated, "Many instances could be given of the good results of this protection, but it seems too obvious to require illustrations, or to be disputed. We will assume then that the advantage of screening, as a preventive measure against malaria is granted."

Such a statement from such an authority on malaria control procedures obviously would convince most observers. However, in the scientific world of today, where proof is ever required to substantiate claims, it does not appear out of order to cite at least one work carefully planned to substantiate the claims made for the benefits produced by this procedure.

Watson and Maher in 1939 conducted such an observation at the Wheeler Reservoir. The charts, appended pages, present graphically the results of this demonstration.

If additional proof of the efficiency of the procedure is needed, an experiment can be planned by anyone in a malarious district. Every other house can be mosquito-proofed, or all houses in one town mosquito-proofed and those in a town with comparable malaria rates left untreated, and the presence of malaria, as measured by spleen and blood examination, recorded and classified over an adequate period of time in accordance with the classification "mosquito-proofed" and "non-mosquito-proofed" houses. However, such an experiment is not necessary and would result in an undue exposure to malaria of those not protected.

L. T. Coggeshall in War Medicine, for May 1943, reports on the results of his studies for the Pan American Airways of Malaria Infection at certain bases in Africa. He states: "Protection of each person against the bite of infected mosquitoes probably was the most effective and practical measure for controlling malaria." "Protection of each person against the bite of infected mosquitoes" would imply the use of such procedures as spray-killing of adults, repellents, protective clothing, mosquito nets, and mosquito-proofing.

It is interesting that under emergency conditions of war the tendency is growing to utilize the personally applied methods together with the inclusion of mosquito-proofing, whereas the permanent measures of drainage and fill are occupying second place.

Such a change of accentuation in the use and application of control procedures is natural when considered from the standpoint of an emergency situation. Under that situation, areas to be occupied cannot, under most circumstances, be prepared in advance for occupancy by drainage. The personnel must be kept free of malaria at any cost. In many instances the area will not be occupied long enough to warrant more permanent means of control than those applicable for the period of the emergency. Time and circumstances furthermore may not permit of a survey of the area to indicate those districts within flight range which are producing the proven vector.

In many instances in the present world war situation, those methods of control which previously were considered accessory to the main methods of drainage and fill are playing, and will continue to play, in emergency situations, the prominent role. They can be made effective, but to accomplish this end all of them must be applied to as nearly as possible one hundred percent perfection. However, it must be remembered that permanent control should still be practiced at permanent and semi-permanent military installations.

Mosquito-proofing has long been an accepted method, classified in the semi-permanent category.

In effecting mosquito proofing, first grade materials and good workmanship are of the highest import. The best grade of cured lumber available in the locality should be secured for the manufacture of the doors and window frames, if the windows are to be screened by the use of frames and wire. The lumber for the doors should preferably be of five-quarter inch ( $1\frac{1}{4}$ " ) thickness to withstand the hard usage for which doors must be designed.

Screen wire should be of a good grade of galvanization. Products from different manufacturers will differ widely in their capacities to resist the action of the elements. Of course, copper screen, aluminum or monel metal are probably preferable, but generally are difficult to obtain. A good grade of galvanized wire will suffice unless exposed constantly to heavy watering with salt water such as may occur near windy beaches at the sea front. The wire should not be less than 16 meshes to the inch, and rarely is it necessary to use more than an 18 mesh wire to prevent the entrance of mosquitoes. The passage of sand flies is a different subject. If an attempt be made to prevent their passage by a wire of small enough mesh to be efficient, it will be found that the ventilating capacity is practically nullified. The subject of plastic screening is not discussed here as it is felt that its recent appearance has not afforded a sufficient lapse of time to judge accurately its exact role as a substitute for wire.

Doors should open outward, and, in localities with a high incidence of malaria, should be protected by a vestibule with a second screen door, also opening outward. Care should be exercised, where space is available, to have the vestibule sufficiently deep so that an individual will be unable to reach both doors at the same time, and hold both open at the same time.

Elsewhere, pages 157, 161, 162, 163, 164, in this booklet is included a sketch of a screen door together with pages for notation of lumber and hardware. By studying the sketch it is believed that anyone handy with the use of tools can make a screen door.

From the utilization of these pages listing materials, it is possible to record the amount of material needed for one house or for hundreds of houses. Especially is this true when, as in the case of military establishments, all similar types of houses are essentially the same. Simple multiplication of the amounts, determined by the measurement as necessary for one house, will determine the total amounts needed.

Windows can be mosquito-proofed by simply tacking the wire on the outside of the frame, or if more sightly appearance is desired, by applying screen wire frames to the windows. When wire is tacked on window frames, a lap over of at least an inch of wire on each side and an inch-and-a-half on top and bottom should be provided.

At times, objection may be raised to the procedure of tacking wire directly on the window frames. Housewives, particularly, will argue that it is difficult to wash windows under these conditions.

There is a procedure which can be used to answer this difficulty. The wire is adjusted so that it covers the window opening with lap-overs as above noted. When in place, the wire is tacked to the top of the window frame. It is then pulled down toward the bottom so that it covers the opening with symmetrical lap-overs. Several tacks are driven partly in to hold it in place. A piece of wood similar to a lath or moulding approximately one inch wide by one-quarter inch thick and cut to the correct length of the window opening is then adapted to the one inch lap-over of wire on the side of the window frame. Starting at the top and being sure that the wire is stretched and not allowed to bulge, brads are tacked through the moulding and the wire underneath the wooden strip, into the window frame at about six to eight inch intervals. The same procedure is repeated on the other side of the frame, at the bottom and at the top as well, if a good appearance is desired.

To remove the wire for window repair or washing, the laths are pried up and the wire may be rolled to the top of the window. If the top of the wire be fastened to the top of the window frame in the same manner, the wire can be completely removed with no damage whatsoever, and utilized in another location where the window opening is the same size or smaller.

Before application of the doors and wire to window frames, the wooden frames themselves on the houses should be inspected to determine if they are in sufficiently good condition to permit thorough mosquito-proofing. When the houses are originally inspected and measured, all necessary odd pieces of lumber should be noted in their proper space on the pages listing materials necessary for such repair work. All such repair work should be performed before installation of screens.

Screen doors and window frames with screens should be applied to the outside of the frame of the house, and no attempt made to fit the door or screen window frame inside of the opening at door or window. This will facilitate greatly not only the work of application, but also simplifies the work of measuring the house, the manufacture of the product, permit re-utilization of the door or frame on another opening of somewhat comparable size, and will result in a great saving of time and labor.

In measuring the doors or windows when they are to be applied to the outside, accurate measurements do not have to be secured. In fact, attempt should be made to make the doors and window frames larger than this open space so that they may be re-used later on larger doorways if desired.

To fix the door or window screen in position once a good balance and correct position is secured, the hanging strip to which the door or window frame is already attached is nailed in place and a moulding strip is nailed to the frame around the door or window screen to serve as a seal around the door or window and insure a close fit through which mosquitoes will not be able to enter. Doors, as noted above, should be made at least two inches wider than the width of the open space of the doorway, and at least one inch longer than the open space of the height. Should the door or screen need readjustment later because of warping, settling of the house, swelling, or other causes, it will be a simple matter, when attached to the outside to make this adjustment.

It is a good practice to paint all lumber before it is cut for manufacture of frames. This insures a better covering with paint than can be effected later after the door is made, and also permits of a neater application of paint. When the door is once in place, it can be touched up with paint in the spots indicated.

Mosquito-proofing, as mentioned above, involves more than preparation of the door frames and their coverage with screen materials. It involves protecting every spot where a mosquito can enter.

The most common spots necessitating such additional protection are the floors, at door and window frames, around the eaves, at the juncture of outside walls, where chimneys and pipes pass through the roof or walls to the outside, openings for pipes and flues and the chimney opening at the hearth inside of the house.

If the floors have cracks which connect with the outside, the floors may be relaid, may be covered with strips of tin or scrap-sheet metal applied either on top or bottom; or strips of wood, comparable in size to lathing, may be nailed over the cracks on the underneath side of the floor. They may also be covered with a covering such as linoleum or oil cloth, or other suitable material.

All available scrap sheet metal should be saved by the malaria control unit for work of this nature.

Walls with cracks can be covered with a good grade of heavy paper such as tar paper that can be pasted or tacked in place. Holes in the walls can be covered in the same manner or covered with pieces of tin tacked in place. Longitudinal strips of tar paper, or suitable heavy paper, cut to an appropriate width, usually one foot wide, can be fitted around the eaves and tacked to roof and walls. When possible to requisition special paper for such purposes, the paper should be equivalent in strength to 90# basis kraft, weighing not less than 1 pound per 34 square feet. Tar paper should be two-ply roofing paper, weighing approximately 35 pounds per 100 square feet. Under emergency conditions, all heavy paper which may have arrived in packing cases, or inside of wooden boxes, or crates should be preserved by the malaria unit for the purposes noted above. This would not represent the material of choice, but could be substituted. Malaria control demands improvisation. Tar paper, or its equivalent, can also be used at the juncture of walls, where openings to the outside may exist.

There is a mixture which can be made by boiling to a homogenous mass soft paper which has been shredded and mixed with flour. Toilet paper serves well in this capacity, but may be replaced by other soft paper where toilet tissue is not available for this work. Exact proportions of the mix can be made and modified in accordance with type of material available. Toilet tissue or shredded newspaper is boiled in a suitable container until a resulting pulp develops. Flour is then added to the extent of one-quarter of the total amount of paper pulp. This is mixed well together, adding a small amount of water if indicated. Two parts sand and one part cement are mixed together and added to this mass of paper pulp just before using, in about the proportions of three to four parts of the pulp and flour mixture to one part of the cement-sand mixture. The resulting plaster can be utilized in locations where other means fail, or when other materials are not available.

It must be recalled that after cement is added to any water mixture, the resultant "mix" will "set" in a short time and accordingly must be utilized quickly.

The opening at the hearths of chimneys may be closed with a suitable wooden covering or a frame and screen covering, or the simple expedient may be adopted of stuffing a bag filled with hay or other suitable materials and shoving it up the chimney so that all openings through which mosquitoes may enter are closed. If the last mentioned procedure be adopted, it will be necessary to remember to remove the stuffing before a fire is lighted.

In association with the subject of mosquito-proofing, it must be remembered that maintenance must be exercised. Buildings must be constantly inspected and provision made for immediate repair by assigning a special person for repair of any phase of the mosquito-proofing which may prove faulty.

All buildings utilized for personnel should be mosquito-proofed, in-

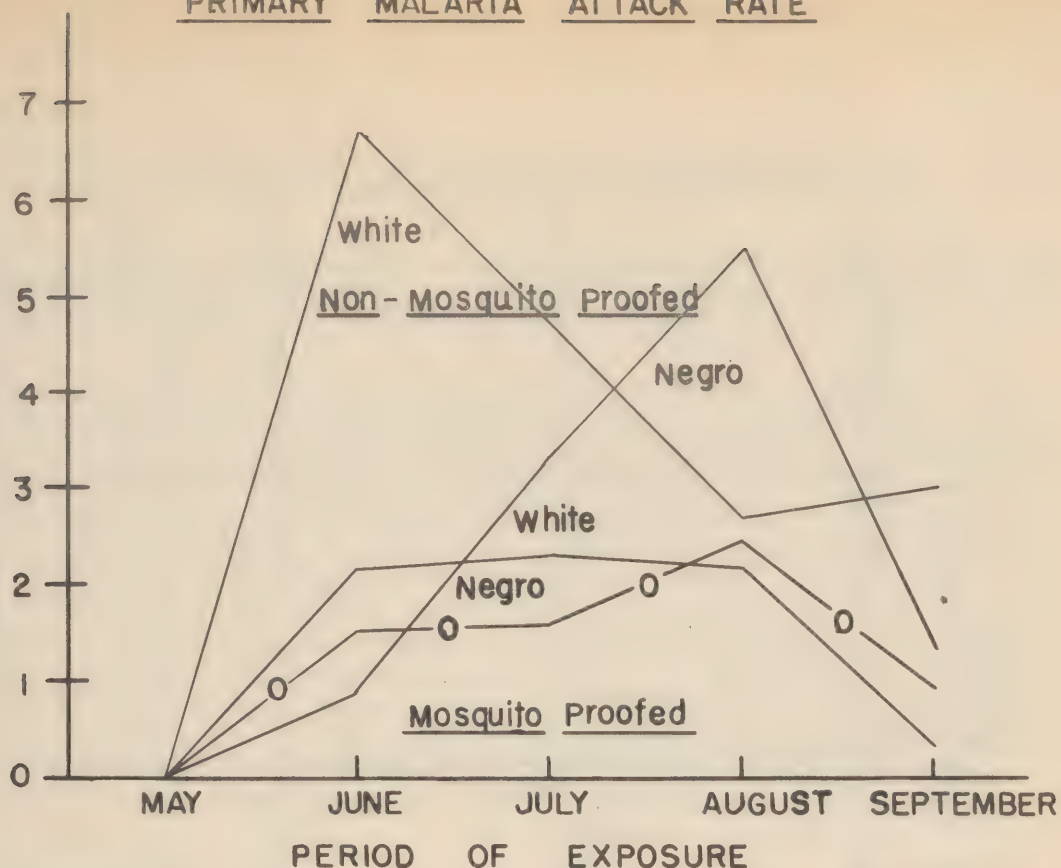
cluding barracks, mess halls, guard houses, patrol houses, latrines, theaters, and all places where personnel congregates.

Repair and maintenance of mosquito-proofing pertains intimately to that phase of malaria control activities known as "malaria discipline".

In the case of military forces engaged in combat, provision should be made at a point back of the front lines where the men may, when combat conditions permit, come for rest and have protection against mosquitoes even if this protection be only mosquito nettings over cots. As soon as circumstances permit, however, mosquito-proofed huts should be provided.

MORBIDITY PER 1000 PERSON-DAYS EXPOSURE

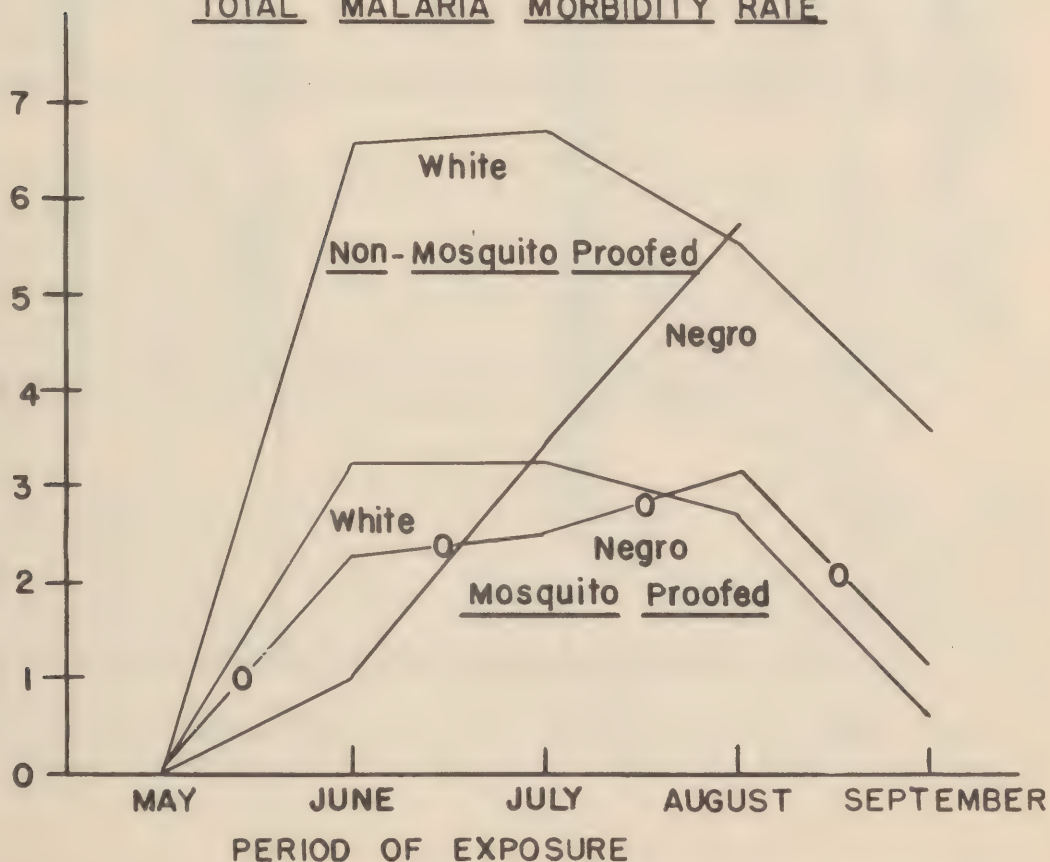
PRIMARY MALARIA ATTACK RATE



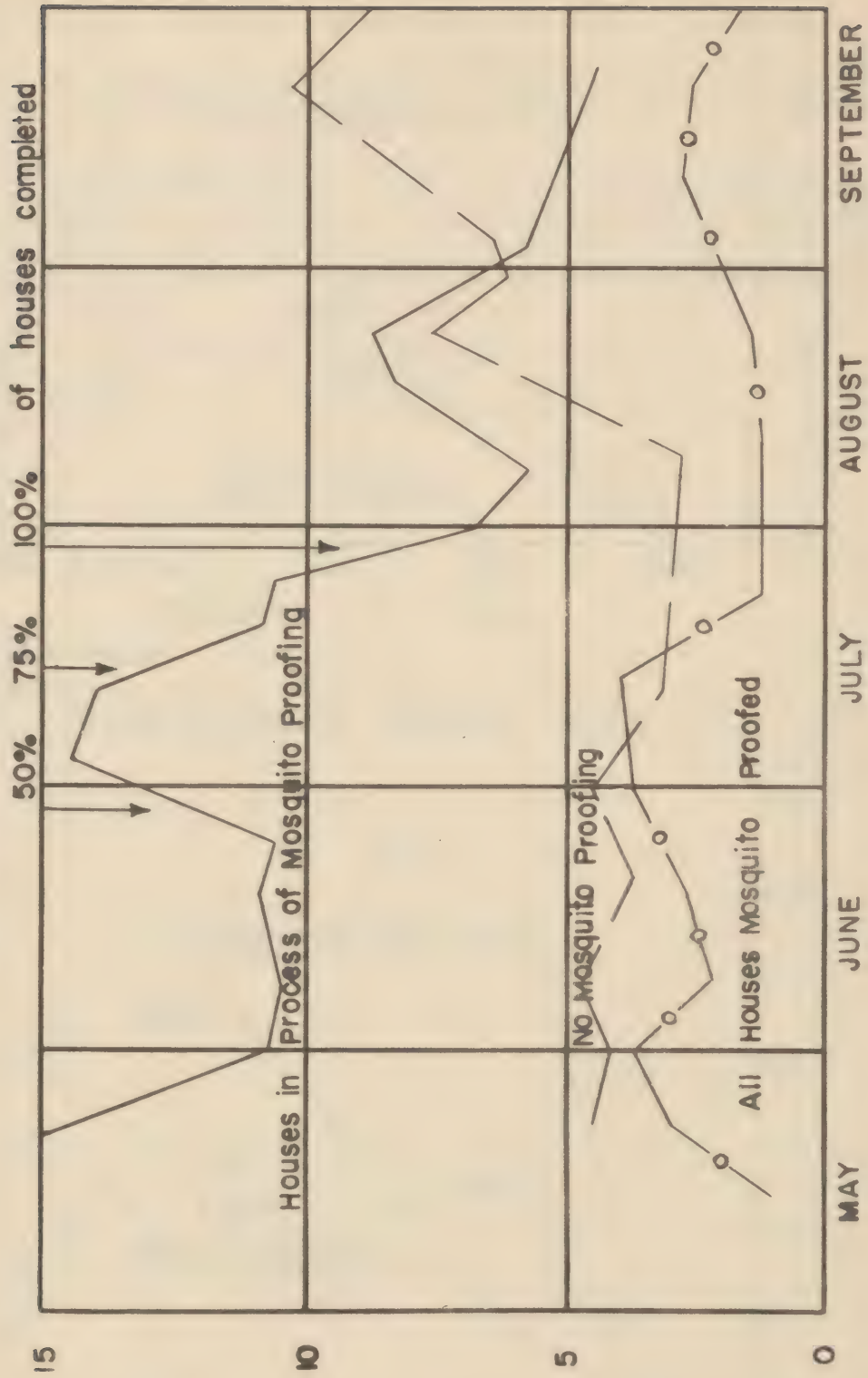
After Watson & Maher-1941

MORBIDITY PER 1000 PERSON-DAYS EXPOSURE

TOTAL MALARIA MORBIDITY RATE



# MALARIA MORBIDITY RATE IN THREE TEST AREAS - WHEELER RESERVOIR - 1939



After Original work from TVA  
Under direction of R. B. Watson

## FLUCTUATION OF WATER LEVELS FOR CONTROL OF ANOPHELINE BREEDING

That fluctuation of water levels has a decidedly beneficial effect on curtailment of anopheline breeding is generally accepted. The exact reasons for the beneficial effect are in some instances still subject to debate.

Several hypotheses, however, may be advanced for explanation of the curtailment of breeding. In the first place, the procedure probably exerts its greatest beneficial influence in the manner of prevention, by producing an environment unfavorable for the breeding of anophelines. The growth of certain aquatic plants results in an environment favorable for production of breeding. When the water is removed by draw-down, these aquatic plants will die. Trees and similar growths, around which debris may collect, are favorable localities for breeding. The removal of these before flooding and impoundment eliminates this factor favorable for breeding. Also, vegetation under the impounded water will have died, due to flooding and, accordingly, draw-down will leave a clear land water edge. At the time of lowering the water level, larvae will be concentrated at the shorelines, and to some extent will be stranded and die. Further, the larvae being drawn down to the new clean shoreline will be unprotected from natural predators.

An additional benefit accruing from lowering the water level is the resultant ease and efficiency with which larvicide can be applied, should this prove indicated, around the clean shoreline resulting from the lowering of the water level. Whether constant draw-down is demanded by the character of the water supply, or whether the intermittent draw-down and refill is practical, the process of procedure and end results are similar.

Certain procedures preparatory to impoundment are indicated. All growth in the impounded area capable of holding debris favorable for breeding should have been previously eliminated. The edges of the high contour elevation of the water line should have been cleared. After impoundment, as soon as breeding starts around the edges, the water level is lowered. Vegetation on the bottom of the reservoir, which was previously covered by water, will have died and the resulting new border will be clear of all vegetation and the factors above mentioned, as prejudicial to breeding, will function until such time as the shoreline vegetation again grows up.

If cyclical fluctuation is possible, the area, after draw-down, can be reflooded approximately to its original elevation after the original lowering of water level has controlled breeding. The water can then be left at this elevation until such time as the advent of breeding would indicate the time again for lowering. Under this situation of constant draw-down and intermittent reflooding with water, it will be found that much vegetation exposed to such a procedure and favorable for breeding will no longer grow.

If refilling be impossible, after the breeding season starts, because of limitations of water supply, the reservoir, after the first draw-down,

would be drawn down again as soon as breeding of anophelines is observed at the waters edge.

Water areas are impounded for various reasons. It may be necessary for power purposes, as reservoirs for drinking water, to answer the possibility of fire hazards, as a source of water for every day use, for swimming, boating, for aesthetic reasons, or even as a hazard on a golf course.

Whatever the reason for impoundment, certain studies should be made and certain precautions taken in areas where malaria is a possible menace before actual impoundment is effected.

In accordance with the volume of water demanded, the height of the dam necessary to provide such supply should be determined. The water shed, permeability of the soil, and average rainfall should be investigated to evaluate the amount of water which can be anticipated over a unit period of time. The contour of the shoreline corresponding to the water level resulting from the height of the proposed dam should be determined. If additional height of the dam is contemplated for the purpose of additional flooding at certain seasons, this resulting shoreline contour should also be determined.

Having determined that the water supply from the water-shed is adequate for the purpose of the impoundment and possible additional flooding, all of the area which will be flooded, as indicated by the contour and topographical study, should be cleared of underbrush and trees. Permanent disposition should be made of the cut trees and brush preferably by piling, and burning. Special attention should be given to the shoreline, as indicated by the shoreline contour, to insure that this edge is clear of vegetation and debris, as well as an additional margin extending beyond this edge, back ten to twenty feet, if circumstances permit. All areas to be exposed during the process of draw-down of water should be graded, and spots of low elevation filled to prevent pooling of water after draw-down.

The area will then be pre-conditioned to a point where potential breeding areas will have been eliminated to the degree possible by advance preparation. It is a recognized fact that larvae propagate better in water areas where vegetation exists, and where floating debris and vegetation can collect around such objects as trees, bushes, etc., as well as in the vegetation at the shoreline itself. Such localities, pre-dilect for breeding, are known as intersection lines, and represent the intersection of water, air, and an object.

Knowing the rate at which water may be impounded and the rate of utilization, it will then be apparent whether sufficient will be available for the purpose of fluctuation, and if so, whether it must be of the slow continuous draw-down type, or whether it can be of the cyclical fluctuation type. If the rainfall or other available water supply, such as rivers, is associated with seasons, heavy for instance at a certain period of the year, and light thereafter, it would be necessary to store water at flood times

to answer the needs during the dry season and permit gradual draw-down.

If the supply of water is plentiful at all times for purpose of refilling, and intermittently continuous, then cyclical fluctuation can be practiced. In this procedure the water level is lowered, and then later raised, if not up to its original height, up to a certain degree of that original level. From the standpoint of malaria control, the period at which this artificial refilling should be performed would depend on various factors, namely, on the condition of vegetation around the then existent water edge, on the period of time necessary for re-growth of the vegetation already exposed by lowering the water level, on the time necessary for killing vegetation while under water, and the status of anopheline breeding in the impounded area.

In association with fluctuation of water levels, the subject of scientific irrigation should be discussed since some basic similarities in the two procedures exist.

It has been demonstrated by various investigators that scientific irrigation, can limit anopheline production as well as, in some instances, produce better crops, especially certain kinds of rice. As the success of malaria campaigns among civil populations will frequently depend upon the good will of the populace, measures detrimental to their interests cannot be indulged in without handicapping the efficiency of the control program.

Scientific irrigation might perhaps more realistically be termed intermittent irrigation, a procedure which bears a similarity on a small scale to cyclical fluctuation. As this name would imply, water is conducted through channels into an area where the irrigation is necessary, allowed to remain for a period, and then later is removed.

From the standpoint of curtailment of anopheline breeding, the period of time over which the water is allowed to remain is dependent upon the length of time necessary for the adult anopheline to develop from the egg stage to adult. If one flooding leaves the area flooded to a degree such that water cannot be removed and all pools dried within the period of the egg to adult development, then intermittent irrigation would be unsuccessful from the standpoint of curtailment of anopheline breeding. When this condition exists, intermittent irrigation should not be practiced, as the prime aim of such controlled irrigation is to prevent or curtail anopheline production while permitting crops to grow normally or even better than under previous conditions. When, under conditions indicated for its use, intermittent irrigation is impossible as an adaptation of control, alternative control measures can, of course, be substituted, such as application of a suitable larvicide, or abolishment of the agricultural pursuit, or installation of an elaborate drainage system especially adapted to the area and designed for the quick removal of water and drying of the land. The last mentioned procedure may be expensive and others of the alternate procedures

suggested may be impossible because of economic reasons or may cause antipathy of the local populace toward the malaria program.

When intermittent irrigation is advised as a control procedure, its selection will depend on the fact that the area can be flooded, the water can be left sufficient time to answer the needs of the agricultural pursuit, the water can be removed, and the grounds dried, before sufficient time has elapsed to realize the cyclical development of anopheline adults. For a justified adoption of the procedure as a control measure, it will be necessary to know the time period involved in the cycle of development of the vector from egg to adult, the water needs of the area from the agricultural standpoint, the speed with which water can be removed, and in accordance with the known porosity of the soil and facilities for subsequent drainage, the period necessary for the soil to dry sufficiently, after the water has been removed, to prevent the presence of standing water adequate for the production of anopheline adults.

A point should always be remembered in association with certain stages of the development of the aquatic stage of the mosquito, namely, that, at times, certain stages of this water cycle can remain alive on a soil sufficiently damp. The possibility of the existence of such a situation can easily be tested by ascertaining if, on reflooding of the area, stages of development of the larvae are found developed beyond the point possible if resulting from oviposition by anophelines after the reflooding occurred.

## FILL

Fill, when employed in malaria control activities, can be defined as material, preferably soil or rock, utilized to raise the elevation of a low lying anopheline breeding area, to an elevation where the standing water will be eliminated, and breeding of the anopheline vector eliminated.

Fill, correctly employed with subsequent grading and drainage where indicated, is one of the most permanent procedures for elimination of anopheline breeding areas and concomitantly one of the best procedures for permanent control of malaria.

There are various kinds of fill. These are customarily classified in accordance with the procedure used to effect the end.

Dredge-fill, hydraulic-fill, and fill-by-dumping, include the types of fill which are most commonly used.

The application of fill for malaria control is greatly simplified, if the agency in charge of malaria control carries records, preferably noted on maps, where breeding areas, actual and potential, are noted, together with an estimate of their size and the type of fill indicated for their elimination. Such notations and entries on maps can be made during the period of routine malaria control activities, be those of the nature of routine entomological investigations, associated with routine or special activities of the engineering staff in the field, or resulting from the information gained by any member of the staff while engaged in other activities.

It is a much more economical procedure if malaria control works can be effected in close association with activities being prosecuted for other purposes. Frequently such related activities will be in progress, and, if the malaria control organization has located breeding areas where fill would be indicated, these areas can be eliminated as a result, or "by-product" of other works and may at the same time dispose of the earth, or fill, frequently in a more economical manner than had originally been planned. Often it will prove impossible to assign personnel and equipment for large projects of malaria control. However, where equipment and personnel are already present for other practical and urgent projects, the equipment and personnel could be, in certain instances, employed for such control over limited periods.

When new construction work is being performed, frequently waste-earth or material will be available, such as after occurs when airfields are being dug. Under such circumstances, if the malaria organization has studied the terrain and determined where fill should be utilized, the waste-earth or material can be "wasted" in areas quite possibly contiguous to, or at least near the point of construction, and result in the elimination of many breeding areas.

Sometimes the necessity will arise for dredging a harbor, lake, or waterway. Those in charge of such an enterprise can, at times, be directed to dump their effluent of earth and water into large areas which were previously anopheline breeding areas. City street-sweepings can be utilized in the same manner for minor or even major breeding areas where fill is indicated as a remedial procedure.

When ordinary refuse is permissible for use as fill for a given area, this material, as well, may be utilized for control of anopheline breeding areas. When utilizing street-sweepings or refuse, precaution should be exercised that the fill be performed in a sanitary manner, that the material used will answer the needs of fill, and will not produce other unexpected and harmful results and that it be performed in accordance with existing sanitary codes of the locality.

In this connection, one instance came to the author's attention where the refuse of a mattress factory was utilized for fill. The greater portion of this waste material was cotton. After being placed in the low lying area, the cotton in some way became ignited and burned for months. Eventually all of the material had to be removed and an earth-fill substituted.

Dredge-fill, sometimes called hydraulic-fill, is, as its name implies, the water and earth mixture resulting from the dredging of the bottom of a waterway. The dredge pumps the soil from the bottom of the waterway and delivers it together with water through large pipes onto the land in the near vicinity. If the pipes delivering the effluent can be placed first at point of the area to be filled, which is to be the resulting point of highest elevation of the fill, the surface of the fill can be self-graded. Starting with discharge of the fill at the point where resulting elevation is the highest, as this area is filled to this elevation, the length of the discharge pipe is adapted to fill (to proper elevation) the succeeding area and so on to the final discharge point. If engineering studies of the area to be filled have been made, and a need for a subsequent underground drainage system is indicated for the drainage of the area itself, or for the purpose of conducting other drainage water through the area, this system may be constructed before the fill is applied to the area and avoid necessity for later excavation for the drain line. Under such conditions, various precautions must be exercised. Indicated elevations of the drain line in the area to be filled must be determined. A firm base for the drainage line must exist, or must be made by laying the tile on previously installed trestles, if the area is boggy. It should as well be firmly held in place laterally by piling to preserve its lateral alignment. Further, the drainage system should be completely blocked off during the period of the actual fill to the end that it does not become clogged with sand.

Hydraulic-fill is the type of fill which produces a deposition of sand silt or other material in a desired spot by application of procedures

involving an understanding of principles of hydraulics. For example, jetties or baffles can be constructed in moving currents of water in order to direct the current or interfere with velocity of flow and deposit material at desired spots. The deposition of material can be determined by a knowledge of water currents, changing of velocity and direction, interruption of flow and the resulting deposition of materials by correct use of these principles. Jetties or baffles can be constructed in a stream flow to direct the current against an obstruction where velocity will be slowed and deposition occur. They can be so constructed that eddies will form behind them with a settling of suspended material. Baffles can be constructed on the bed of a stream in such manner that, given correct resulting velocities of flow, suspended materials will be deposited below these baffles and the stream bed raised. Simple changing of a gradient from a greater to a lesser grade can be utilized, under the correct relationship of the two gradients, to produce deposition of material.

A current of water entering an area of large surface will be slowed and deposit suspended material. In drainage systems where a large amount of sand is encountered, ditches can be constructed to utilize this principle. The water and sand can be conducted by means of specially constructed canals into a large area needing fill. The slowing of the water when smaller conduits discharge into this area of large surface will, as noted above, cause the suspended material to settle out. When the deposition of the water borne material has raised the level of the elevation of the area, the resulting accumulated water may be drained from the district by a drain located at a suitable low point.

Dump-fill, as the name implies, is fill which is transported to the area by any means and dumped in place.

Fill should usually be graded after it is in place to prevent the formation of surface pools. Tractors, wheelbarrows, horse drawn slips (or scoops), tractors with graders, bull-dozers with graders may all be utilized to accomplish this end.

After filling has been performed, the area should be watched for shrinkage, for cracking, as well as for lowering of elevations of the surface. Because of these characteristics of new fill, it is unwise to install drainage lines until all such change in configuration due to shrinkage has occurred, unless as recommended above, the line is held in place laterally and vertically by adequate support. Fill, well graded and well executed, can eliminate anopheline breeding areas permanently and, of course, frequently reclaims valuable land.

## TIDE GATES

Tide gates are frequently of use as an adjunct for the elimination of water areas in which anophelines may breed. They are most commonly employed as a means of prevention; that is, to prevent water from occupying a low lying area, or from flooding an area at excessively high tides with the resultant formation of pools and water areas in which anophelines may breed.

As the name implies, they are used where tides exist, or where the raising of the water level may occur from other causes than the influence of tides. Among such other causes for the raising of water levels would be such situations as freshets in rivers, floods recurring frequently or at stated seasons.

The ideal location for the effective function of a tide gate is one where the shoreline is of relatively high elevation, and where areas of lower elevation exist through which storm flood water or seepage water must be discharged into the body of tidal water, and where the area to be drained could be drained were it not for the stoppage of the system or the back-flooding of lands by the advent of high tides.

For the drainage of an area presenting these conditions, an ordinary and adequate drainage system should be planned. The average volume of water to be discharged through the system over a twenty-four hour period should be computed; the height to which the water level of high tide would rise in the system and cover the land basin involved should be determined; and the degree to which the drainage of the system would be hampered during the period over which the tidal high-water level remains should be computed. The elevation of the proposed drainage line should, of course, be known, as well as the total period during twenty-four hours when the gate will be closed due to high tides.

With these facts in hand, it can then be computed whether a tide gate is essential for adequate drainage of the area, and if so, what other elements of a drainage system must be incorporated in the plans to make drainage adequate and successful.

With the average difference between water elevation at high and low tide known, the resultant expressions of this water level throughout the drainage system at high tides can be determined. If the outfall point is at the elevation of average low tide, and the tidal differential is two feet, it is obvious that the rise in water elevation of two feet at the outfall will back-flood through the discharge system up to the two foot contour of the area to be drained. All contours, or land elevations, lower than two feet, would theoretically receive an additional volume of water at high tide, entering the area in a direction reverse to the ordinary flow of the system at low tide. Under these conditions, the only portion of the system from which surface water would be removed, and in

which ground water levels could be lowered, would be that portion of the system where water head would be above the two foot elevation. Under the above circumstances, during the period of high tide, the portion of the system, up to two foot elevation, would function as a conduit only for the water drained from the area above the two foot elevation.

A tide-gate is indicated when the volume of combined water, tidal and run-off, which is introduced into the area, is such that discharge is incomplete, during the period of low tide, with the resultant presence of residual waters. If the volume of the water is such that it can all be discharged during the period of low tides, then no tide gate is necessary.

If the ideal situation of a shore line elevation exists, higher, except at natural discharge points, than the high tide elevation, the problem of utilization of a tide-gate is simplified. When such an elevation does not exist, it is sometime necessary to construct a dike or wall to hold the high tide-water from flooding the area.

A tide-gate is a device hinged at the top which hangs in front of the outfall pipe or canal and can close the outlet of the drainage system or pipe. When the tide rises, the water pressure on the outside of the gate becomes greater than the pressure of drainage water inside, and the gate closes. Outside water then cannot enter the system either through the discharge drainage canals or over the top of the shoreline, which has been diked, if necessary. During the period of closure, water from the drainage system naturally accumulates inside the system and exerts pressure on the inside of the gate. As the tide falls, a point is reached where the inside pressure is more than the outside pressure, the gate swings open, and the accumulated water inside the system is discharged. During the period of closure, the system has functioned as a drainage system, unobstructed by the entrance of outside water into the system. However, it has not functioned at its greatest efficiency as drainage has been progressively more impeded by the accumulation of its own waters at the temporarily closed outfall end of the system.

At times, when the water shed to be drained is extensive and the volume of drainage water large, water will be accumulated throughout the system, when the gate is closed, because of no discharge point at the lower end of the system adequate in surface area to reserve and temporarily store the waters. Under these circumstances, a storage reservoir, will have to be constructed near the outfall end of the line to store the water from the upper reaches of the system during the period when the gate is closed, and which, of course, will be discharged when the gate opens. The size of such a reservoir would naturally be governed by the estimated volume of water to be temporarily stored and the resulting elevation of this stored water. The greater the surface area of the reservoir for a given volume of water, the lower the elevation of the resulting accumulated waters.

This reservoir will provide a temporary point of discharge for this drainage water and its temporary storage at the lower end of the system,

and, allow the system to function to a degree approaching the drainage capacity of the system when the gate is open.

Gates may be employed in special manners. Special situations exist where it may be desired, for example, to hold high salt water tides in a lagoon otherwise filled with water sufficiently fresh to permit of breeding of anopheline mosquitoes. Tide gates mounted to open inwards can accomplish this end automatically. However, under such circumstances, if personal attendance is possible, it is better to use a manually operated gate. Utilizing this procedure, drainage, when volume of water is normal, can proceed from the area in question, and when a high tide of salt water enters the area, it can be held there by lowering the gate, and a breeding area of fresh water anophelines may be rendered sufficiently saline to prevent breeding.

In association with the utilization of gates, another use should be mentioned. In this instance, they would be used to permit of the process already described and known as draw-down fluctuation. When a breeding area exists near tide water, either fresh or salt, high water levels, resulting from particularly high tides at certain seasons or due to flood water of any nature, could be held inside the area, flooding it to an elevation higher than normal; then, as indicated, draw-down fluctuation could be practiced.

## USE OF DYNAMITE IN MALARIA CONTROL

### 1. INTRODUCTION

Dynamite has long been recognized as an economical means of ditching under favorable conditions. In recent years, because of scarcity of machines and hand labor, it has been used more and more on ditch construction. It offers a maximum of expendible energy in a minimum of bulk. Its use is relatively safe if reasonable precautions are taken, but its use, to be effective, requires careful study and good sound judgment in each project attempted.

Dynamite is defined as a high explosive which decomposes almost instantaneously by detonation. There are several types of dynamite in use, each of these having been subdivided into different grades or strengths. Straight dynamite or nitroglycerin dynamite, as it is commonly referred to, is a mixture of nitroglycerin and other substances, some of which are also energy producing. Ammonia dynamite is so named because the amount of nitroglycerin is reduced and energy producing ammonium nitrate added. This resulting mixture is the base explosive of ammonia dynamite although other energy producing products are combined to make the final product. Gelatin dynamite takes its name from the gelatin, a jelly-like mass of soluble nitroglycerin, which substance is used as the basic explosive. Other substances, some of which are energy producing, are utilized to form the commercial product, gelatin dynamite.

Dynamites possess several characteristic properties which bear a definite relationship to the determination of the amount and type of explosive needed for specific uses. First, the term "strength" refers to the energy content of dynamite. The straight or nitroglycerin dynamites are rated according to the percentage by weight of nitroglycerin they contain. For example, a straight 20% dynamite contains 20% by weight of nitroglycerin and 80% other substances. It does not follow that a 40% straight dynamite is twice as powerful as the 20%. Even though nitroglycerin has been increased 100%, other energy producing compounds have been reduced 25%. Such an erroneous concept of dynamite strengths is wide-spread among laymen.

Ammonia dynamite and gelatin dynamite are rated in the same manner, per cent by weight of their basic explosives. Their bulk is also supplemented with other energy producing substances.

Density of dynamite is expressed as the number of  $1\frac{1}{4}$ " x 8" cartridges in a 50 pound box. Varying densities are desirable so that greater flexibility in loading is possible.

Water resistance of dynamite is quite important since blasting of channels is often done in muddy soil, and even under water. Gelatin dynamite

mite is practically waterproof, nitroglycerin dynamite is resistant to moisture especially in high density grades, and ammonia dynamite can be used under water only in the higher densities.

Velocity as pertaining to dynamite is the measure in ft./sec. of the detonation wave that travels through a column of explosives. As velocity is increased, detonation is more rapid, thus producing greater shattering effect.

Resulting gases or "fumes" from a dynamite explosion vary with the type and strength of explosive. Principally these gases are steam, carbon dioxide, nitrogen, and three others which are present in lesser degrees and which are poisonous, carbon monoxide, hydrogen sulphide, and nitrogen oxides. It is not likely that any of these gases will be concentrated in open areas in harmful amounts. However, the well known effects from slight amounts of carbon dioxide will be noticed if personnel walk into the "fume" before allowing it to disperse.

## 2. PROPAGATION DITCHING

Of all the types and varying strengths of dynamite, the one of particular interest to malaria control engineers is 50% straight or nitroglycerin dynamite. This product has been developed by DuFont after much research. It combines all the desirable characteristics of a ditching dynamite. It is relatively waterproof, has high velocity and density, will propagate more easily than other types of dynamite, and, of particular interest to malaria control engineers, it produces a shattering effect which blasts side slopes at an approximate 1-1 slope.

Until recently the great majority of dynamite ditching was done only where propagation was possible. Propagation is simply the impulse transmitted through the earth which detonates the charges along the line to be blasted when charges are placed at a distance of 12 to 24 inches apart. The first charge must, naturally, be set off by means of a cap or fuse. Sufficient force travels from charge to charge and detonates the entire line of charges almost instantaneously. Unfortunately, propagation is possible only in water logged soil. Such a condition momentarily seals the gases caused by explosion and thereby diverts sufficient force horizontally to the next charge to cause detonation. An exploding charge of dynamite exerts equal forces in every direction, but naturally, the majority of such forces will tend to seek the path of least resistance, upward. Soil, not sufficiently saturated with water, is not susceptible to propagation. Under such conditions, a cap for every charge is necessary. This method of blasting will be discussed later.

No set rules can be devised for propagation blasting. Amount of dynamite per cubic yard of material, depth and distance between charges, and indeed, whether propagation is possible at all, can be demonstrated only by setting off a few trial shots. However, past experiences indicate that

a few "rule of thumb" generalities can be mentioned. Ordinarily, one pound of 50% straight dynamite will remove one cubic yard of soil. Propagation is usually possible if the soil contains enough moisture so that a pinch of it, held between the thumb and forefinger and mashed, will yield water. Charges are placed to a depth of approximately 50% of the depth of the desired gradient. Charges are placed from twelve to twenty-four inches apart, certainly never farther. The above generalities can be used simply as a basis for making test shots. Only very experienced blasters are sufficiently experienced to calculate charges, etc., without the aid of test shots. Certainly a relatively inexperienced blasting crew should resort freely to the use of trial shots. Soil type, specific gravity of soil, and moisture content are a few of the changing conditions encountered in various localities. It can be said that most inexperienced blasters have a tendency to set their charges too deep. The more moisture a soil contains, the shallower the charges are set. In very wet muck, the top cartridge may be within 4" of the surface. Soil containing only enough moisture to propagate will require charges being set deeper. At no time should the top of the last cartridge loaded be lower than 12" from the ground surface.

As has been stated, a ditch blasted with 50% nitroglycerin dynamite usually has a 1-1 side slope. The top width is usually equal to twice the depth, plus the bottom width, which is from one-third to two-thirds the depth, depending on soil conditions. With knowledge of this information the engineer has only to stake out the proposed ditch line, run differential levels, and, from field notes, plot a profile. Since the shape of the ditch is already known, and the depth from any point can be taken from the profile, it is relatively simple from there on to figure the charges needed in each hole. In computing cubic yards of earth to be removed water volume overlying the ditch line should be figured the same as if it were soil.

In water logged soils, holes can be punched with an iron crow bar, a sharpened broomstick, or more elaborate punches, such as the Ashley core punch. Loading should be done by the "column" method in propagation ditching. This is merely the placing of cartridges on top of each other, end to end. The remainder of the hole is filled or "stemmed" with soft mud and packed with a wooden or bamboo pole, never metal. If water fills the hole, no further stemming is necessary.

Theoretically, there is no limit to distance over which charges will propagate. However, due to possibilities of varying soil conditions, it is best to fire the line of charges before it is longer than 400'-500'. Needless to say, for safety reasons, the "primed" cartridge or the charge with the cap in it is placed at one end of the line of charges.

### 3. PARALLEL LINE AND CROSS SECTION METHOD

Where wide shallow ditches are desirable, the blaster uses what is

known as the "parallel line" method or the "cross section" method. Accompanying illustrations of these methods are self-explanatory. Charges can be figured on the basis of information already given.

#### 4. POST HOLE METHOD

The post hole method of loading takes its name from the post hole auger, the tool commonly used to drill holes for the charges. Primarily, this method of ditching is one adapted to soils which are too dry to allow propagation, but must have a primer in each charge and be fired electrically. This method of loading can be used in propagation however. Experience indicates that less dynamite per cubic yard is used by the post hole method than by the column method on ditches six feet deep or more. The post holes are drilled to approximately two-thirds of the required depth of the ditch, and from three to six feet apart. Charges are placed in bundles as shown.

One cap for every hole is necessary when firing electrically. Earth is thrown in, in layers of two or three inches, and tamped firmly with a wooden pole, until the hole is filled.

The protruding wires from the caps are connected, preferably in series as shown in the accompanying diagrams. Lead wires are then connected to the free wire on either end of the series, and these are connected to the source of current after all holes are loaded and tamped and the area is cleared.

Admittedly, the blasting of a ditch through dry soil where holes are difficult to dig is a costly procedure. The cost of digging the holes, time consumption of placing a cap in each hole, and the cost of the caps themselves makes electrical firing much more expensive than propagation. Using the post hole method of loading and firing electrically may be a more expensive method of ditching than with a drag line or by hand labor. However, under emergency conditions, if neither labor nor drag lines are available and dynamite is, there should be no question regarding its use in malaria control projects. Especially, is this true in emergency situations where immediate protection of our armed forces is needed.

#### 5. ADVANTAGES OF DYNAMITE DITCHING

As already mentioned, dynamite offers a maximum amount of energy in a minimum of bulk. Thus, transportation of energy producing equipment is minimized and moving sufficient quantity for the task assigned even over unfavorable terrain is relatively easy as compared to difficulties of moving heavy equipment to accomplish the same end. In ditching with dynamite a minimum of right-of-way clearing is necessary. Drag line ditches must be cleared 30'-50' on one side to allow passage of the machine. This clearing introduces sunshine which encourages breeding of many species of anophelines. Too, the movement of heavy machinery over boggy areas often is most difficult. Such movement leaves indentations in soft

earth which may later prove to be prolific anopheline breeding places. One of the most popular advantages of dynamite ditching is the fact that disposition of spoil is taken care of in the initial operation. Propagation ditching is more rapid and usually costs much less than any other available method of ditching.

Some disadvantages of ditching with dynamite are obvious. Its use is considered to be associated with a certain degree of danger. The finished ditch is rough and often requires some shovel work for completion. Some engineers object to dynamite ditching because they wish to utilize the spoil, which is impossible when using dynamite. However, if all these objections prove unimportant, dynamite ditching will prove to be a valuable asset in temporary malaria control projects.

## 6. OTHER USE OF DYNAMITE IN MALARIA CONTROL

### a. CLEARING DITCH LINES

When huge boulders, stumps, or logs are encountered in a ditch line, a high velocity dynamite can be used to remove them. The accompanying diagram shows methods of loading for boulder blasting. This should be a separate operation in itself. In the case of stumps and logs, their removal can be accomplished at the same time the ditch is blasted. Extra charges are simply placed under these obstructions.

### b. CLEARING LOGS AND STUMPS FROM STREAMS

Stream larviciding is often done from a small boat because of high banks and thick vegetation on the shores. Where fallen logs have blocked the stream channel, and boat travel is prohibited, such log jams can be easily removed with dynamite. At the desired point of breakage an auger hole is drilled in the log and the charge placed in this hole. Logs and stumps under water can be cut in two by making a noose of dynamite sticks and placing this around the desired point of breakage.

## 7. DISCUSSION OF BLASTING CAPS

Thus far caps have only been mentioned casually. There are two general types of caps, those fired by a safety fuse and those fired electrically. Caps are small charges or detonators which are exploded to induce detonation of the basic charge. A blasting cap consists of a small copper tube approximately  $1\frac{1}{2}$ " long and  $\frac{1}{4}$ " in diameter. This tube contains a high velocity explosive, tetryl, which is also highly sensitive. The caps are fired by an electrical current. Two insulated copper wires lead to the explosive, tetryl, inside the copper tube. The wires are held in place by a wax like substance. A current is passed through the wires to cause the explosion.

Caps fired by safety fuses are comparable to large firecrackers in

construction. This type of cap has an opening in one end which the fuse fits exactly. A sufficient length of fuse is cut off, the cut being square, perpendicular to the length of the fuse and is inserted in the opening of the cap. Then the cap is crimped so as to hold the fuse in place. A special crimper is designed for this purpose and pliers or use of teeth, as is sometimes seen in cases of careless blasters, should not be substituted for it. Safety fuse is waterproof and will burn under water. Rate of burning in ft./sec. is stamped on the carton. Usually this is from 1 ft./30 secs. to 1 ft./60 secs. Caps and fuses are always brought separately and loaded as needed.

The handling of dynamite alone is not particularly dangerous until caps have been loaded or the charges have been "primed". Caps are highly sensitive and therefore most dangerous. The mere explosion of one cap may delete several fingers or destroy a person's eyesight. Caps must never be stored or transported in any manner along with dynamite. Too much care cannot be exercised in the handling of caps. Past records indicate that most accidents in handling explosives have been from careless handling of caps.

#### 8. BLASTER'S ACCESSORIES

The blaster has several machines and tools, some of which have been mentioned, with which he works. Only the common and necessary ones will be noted here.

Blasting machines are used to generate current when firing by electricity. They are simply magnetos or modified generators which produce current when certain mechanisms are operated. The rack bar type, which is a modified generator in effect, produces a pulsating direct current when the rack bar is raised and pushed down very rapidly. The machine consists of two field magnets and a revolving armature which are arranged in the usual fashion of generators. It is so designed that no current is released until the end of the stroke of the rack bar. The current is released then to the firing line. These machines are rated in accordance with the number of caps they will fire simultaneously, as 30, 60, 100. Probable resistances of lead wires have been taken into consideration in determining these ratings.

The circuit tester, erroneously called a galvanometer, is a small machine which consists of a chloride of silver dry cell connected first to a coil, between the poles of a magnet, which actuates a needle mounted on delicate bearings, and then through a high resistance coil to two poles. When a circuit is connected to the two poles, the needle moves across its scale according to the resistance encountered. No movement of the needle indicates an incomplete circuit; slight movement indicates much resistance; while much movement means very little resistance. The circuit tester is used to locate faulty connections in a circuit.

The rheostat consists of five coils of wire of known resistances which

are connected with six binding poles in such a manner that the resistance between any two posts is constant. It is used for testing the efficiency of a blasting machine.

Crimping pliers are used on the cap which requires a safety fuse. A slight crimp is made in the cap at its outer extremity for the purpose of holding the fuse in. Ordinary pliers should never be used. Standard crimping pliers are so shaped that a cap fits snugly in the slot provided and allows equal compression from all sides.

The blaster should construct wooden tools for use with dynamite since metal tools should never be used because of the danger of making sparks. A minimum of three tools, a wooden maul, a wedge for opening boxes, and a punch for punching primer holes in dynamite sticks, should be provided.

Rubber gloves are worn by some blasters in an effort to prevent headaches. The nitroglycerin in dynamite, coming in contact with the skin, causes a lowering of blood pressure and a resulting headache.

Other accessories may be used, such as the Ashley core punch for punching holes, but, in general, very little equipment is needed for blasting.

## 9. SAFETY PRECAUTIONS

Handling of dynamite is usually considered to be hazardous. However, if a few rules are followed religiously, and common sense is used, there is no cause for fear. The blaster must never acquire a nonchalant attitude, but must always have a respect for the enormous power in dynamite and caps. Following are a few of the don'ts regarding the handling and use of dynamite. These are taken from a list published by the Institute of Makers of Explosives and are the result of numerous accident reports for many years.

- a. Don't store blasting caps and dynamite in the same magazine.
- b. Don't smoke while using or handling explosives.
- c. Don't store explosives so that cartridges stand on end.
- d. Don't store dynamite or caps in a wet or damp place. They should be kept where it is clean, cool, dry, and well ventilated.
- e. Don't transport dynamite and blasting caps together under any circumstances.
- f. Don't handle packages of explosives violently or slide them roughly along floors.

- g. Don't handle explosives near open lights, other fire or flame, or sparks.
- h. Don't use any tool except wooden ones in opening dynamite cases or for stemming.
- i. Don't leave explosives or waxed paper that has been wrapped around explosives where stock can get at them. Cattle and other stock like the taste of salts in explosives, but these or other ingredients will probably make them sick or kill them.
- j. Don't use damaged leading or connecting wire in blasting circuits.
- k. Don't fire a blast before all persons are at a safe distance or under adequate cover, and all explosives are removed a sufficient distance and in a safe place.
- l. Don't attempt to investigate a misfire too soon, even though it is thought that the fuse has not been lighted or has gone out.
- m. Don't leave packages of explosives or blasting caps uncovered or exposed to the direct rays of the sun.
- n. Don't carry blasting caps in the pockets of your clothing.
- o. Don't attempt to investigate the contents of a blasting cap.
- p. Don't allow priming to be done in the magazine.
- q. If using blasting cap and safety fuse, don't:
  - (1) Cut fuse on slant, but square across.
  - (2) Cut fuse too short.
  - (3) Crimp caps with anything other than cap crimper.
- r. Don't lace fuse through cartridge of explosives.

## APPENDICES



## PREPARATION OF BLOOD FILMS

The preparation of blood films for both clinical and survey studies is a simple technical procedure that is often incorrectly performed. A film worth the taking is worth taking and staining well. "... Well made films are the basic step toward reliable diagnosis and the best efforts of a qualified technician may be frustrated by the poor condition of the material with which he has to work." The conscientious and routine application of the following principles will produce films of good quality and thereby eliminate many of the pitfalls of routine examination.\*

### 1. THE SLIDE MUST BE CLEAN.

A blood film placed on a dirty slide frequently washes off during the process of staining. Dirt takes up stain and the number of artefacts in the film is greatly increased. There may be acid or alkali deposits on the slide that will interfere with staining by changing the pH of the stain. New slides must be cleaned before use. The following method is recommended:

- a. Wash in warm soapy water.
- b. Rinse thoroughly in running tap water.
- c. Rinse in distilled water (if available).
- d. Dip in ethyl alcohol (90-95%).
- e. Dry with a clean lintless cloth.
- f. Protect from dust and dirt.

Care must be exercised in handling slides after they are clean as grease or dirt on the fingers will cause thick smears to slip off. Always handle clean slides by the edges.

### 2. CLEANSE THE SKIN THOROUGHLY.

Dirt, grease, bacteria, etc., may be removed from the area to be punctured by cleaning with gauze or cotton soaked in alcohol. Then dry the area with clean, sterilized gauze, or cotton, since alcohol mingling with the blood will fix the red blood cells, and interferes with staining because of faulty dehemoglobinization.

### 3. PRICK THE SKIN DEEPLY.

A deep prick through the skin will cause enough blood to flow without having to apply undue pressure. An instrument with a cutting edge is usually most efficient. Sharp pointed surgical blades (#11, Bard-Parker), hypodermic needles (#23 or #24), pyramidal pointed needles and pen points have all been used successfully.

\* N.I.H. Bulletin #180, U.S.P.H., Aimee Wilcox, page VII, Preface.

#### 4. MAKING THE FILM.

##### a. THIN FILM

Place one or two small drops of blood near one end of the slide, parallel to a line corresponding to the slide's width. Place the slide on a flat surface, and incline another clean slide over it at an angle of  $30^{\circ}$  with the horizontal. Slip the upper inclined slide on the lower one, holding the slide on which the drops of blood rest until the drops touch its underside, and coalesce along the point of contact of the two slides. Then with a rapid motion, push the inclined slide to the far end and the thin film will be spread thinly on the slide below. It is important that the blood should be pulled across the slide, and not pushed ahead of the inclined slide. Allow the film to air dry.

##### b. THICK FILM

The thick film should be about the size of a dime and placed near one end of the slide. Touch the under surface of the slide to the drop of blood on the finger or ear and move in a circular manner until a smear of desired size and thickness is obtained. An alternate and equally efficient method is to take two or three drops of blood of average size, closely grouped on one end of the slide, and puddle them to desired size with the edge or corner of a clean slide or glass rod. Place the slide on a level surface and allow to air dry.

If the resultant film is too thin, the advantages of the thick film method will be nullified; if too thick, the film will crack and peel. Ordinary printing should just be read through a properly made wet thick smear.

Thick and thin films may be combined on the same slide. The thin film can be stained, when desired, and used for detail study.

#### 5. IDENTIFY THE SLIDE.

Use some type of marking either name or number, and have a date sheet to correspond to this identification. The name or identifying mark may be written in a thin blood film with a pencil or pen. This, however, cannot be done in a thick film as the film will be loosened, and then subsequently lost in the process of staining. A colored wax marking pencil is quite satisfactory.

#### 6. PROTECT THE FILM.

Cover the freshly made smear with a petri dish or place the slide in a slide box, standing on end, film side down. Keep the film protected from insects, especially flies and roaches, as they will rapidly eat all of the blood off the slide.

7. STAIN AS SOON AS POSSIBLE.

Films may be stained a few hours after they have been procured. If they are allowed to go unstained for too long, i.e. weeks and months, the film may harden to the point that dehemoglobinization will be very difficult, and usually incomplete.

## THICK BLOOD FILM DIAGNOSIS

The thick blood film was devised as a means of examining a relatively large amount of blood in a short period of time, thereby obtaining a more rapid detection of the parasite. This, of course, increased the percentage of positive examinations and effected a closer correlation of positive laboratory examination with clinical and survey studies. Instead of examining individual RBCs the cells are heaped up in layers and the hemoglobin of the cell washed out in order that light may be transmitted.

Inasmuch as the outline of the RBCs are obliterated, the microscopic picture differs considerably from that of the thin film. The background varies from a grayish-blue or light blue in the thickest portion to a pink or clear field along the thinner periphery, all depending on several factors such as the age and type of stain, length of time in staining, how long the film has dried, etc. The nuclei of the WBCs are purple and platelets stain a pinkish-violet. The outline of the RBCs usually cannot be seen except in the thinner portions of the smear where they appear as "Ghost Cells".

The parasites appear slightly smaller and shrunken, and often do not have their characteristic shape. Portions of the cytoplasmic structure are frequently broken off and granules, at times, do not stain.

Species diagnosis is possible in a thick film, but is often very difficult, even for an expert malarial parasitologist. If there is any doubt regarding the species, always include that in the report. Staining of the thin film at the other end of the slide, if the two types of smears were originally made, will assist greatly in identification of species. Here are a few characteristics that will be helpful in their differentiation:

VIVAX - The presence of a parasite identified as an amoeboid form is virtually diagnostic. All stages are usually found, although ring forms may predominate. The vivax ring is usually considerably larger than those of falciparum. The cytoplasm is usually incomplete giving rise to descriptive terms such as the "swallow form", "comma", "exclamation mark", "interrupted ring", etc. Mature and presegmenting schizonts are similar to those seen in a thin film; although, they may be distorted in shape.

MALARIAE - All forms are usually present and this parasite is more likely to retain its shape than the other species. The forms appear even more compact than in the thin film, and the pigment is prominent. The mature schizont with its eight (8) merozoites can be clearly distinguished and is diagnostic of the species.

FALCIPARUM - Here again, as in thin film examination, there exists the diagnostic feature of finding only ring forms and/or gametocytes. The latter may be readily identified by their crescentic shape. The rings are small

and delicate, the cytoplasmic ring rarely complete, and they are usually present in much larger numbers than the other species.

## STAINING

### 1. PREPARATION OF STOCK STAINS

Stain in the dry powder form is available as a regular Medical Department Supply Table item. Glycerin and methyl alcohol are similarly supplied. An evacuation hospital or any larger medical unit has the necessary technical equipment to prepare stock stains. A survey unit, however may find it difficult to prepare stains under field conditions. The basic materials and reagents necessary to prepare stock stain can be taken to such a unit, and compounded with their equipment. Usually a high degree of cooperation will exist, and material will be exchanged for already prepared stock stains. An ample supply of well prepared stain should be kept on hand at all times; especially, prior to proceeding into a new area where, perhaps, facilities for manufacture may not be available.

#### a. GIEMSA STAIN (ACCORDING TO TM 8-227)

Giemsa Powder . . . . .	0.5 Ggm.
Glycerin . . . . .	33.0 c.c.
Methyl alcohol (absolute, acetone free) . . . . .	33.0 c.c.

The powder is dissolved in the glycerin for one-and-a-half to two hours in a constant water bath at 60° C. The alcohol is then added, and the stain allowed to stand for a day or more before using.

#### b. WRIGHT'S STAIN (ACCORDING TO TM 8-227)

Wright's Stain Powder . . . . .	0.3 gm.
Glycerin . . . . .	3.0 c.c.
Methyl alcohol (absolute, acetone free) . . . . .	97.0 c.c.

The stain and glycerin are ground together and the alcohol then added. A day later it is filtered and set aside to age.

### 2. DILUTION OF THE STOCK STAIN

The dilution of the stock stain is probably the most important step in staining. The pH of the diluent should be in the range from 6.8 to 7.2. A specially prepared solution buffered to a pH of 7.0 with disodium hydrogen phosphate (anhydrous) and potassium dihydrogen phosphate (anhydrous) will give the best results. The solution must be covered with a film of oil to protect it from the air, which will change the pH. (The preparation of this is described in TM 8-227). For general use, neutral distilled water is the best as it usually is available, and also is easy to maintain under field conditions. Tap water may be used if the pH is correct. The Brom-thymol-blue pH indicator has a good range of differentiation around the neutral point. If one is not available, a set may be borrowed from the Engineering Unit supplying water in the area, or from an Evacuation Station

or General Hospital.

The choice of diluent, (tap water, distilled water, or buffered water), will depend upon their availability and the problems of their preservation. If the pH is properly and constantly controlled, the parasites will be well stained.

### 3. SPECIAL STAINS - FIELD'S STAIN

The British are now routinely using Field's stain for their thick film work. The advantages of this method are two. First, the speed and ease with which staining is effected, and, secondly, the conservation of material for the stain may be used over and over again.

#### a. Preparation of stain:

##### (1) Solution A:

- (a) Methylene Blue (medicinal) . . . . . 1.3 gm.  
or (Methylene Blue - 0.8 gm.)  
(Azure 1(B) - - - 0.5 gm.)
- (b) Disodium hydrogen phosphate (anhydrous) . . . . . 5.0 gm.
- (c) Potassium dihydrogen phosphate . . . . . 6.25 gm.
- (d) Distilled water . . . . . 500.0 c.c.

The dye and the disodium hydrogen phosphate are dissolved in 50.0 c.c. of water, boiled and evaporated in a water bath almost to dryness. This residue, plus the potassium dihydrogen phosphate, is dissolved in the water and set aside for twenty-four hours. The solution is filtered after standing for twenty-four hours, and is then ready for use.

##### (2) Solution B:

- (a) Eosin (yellow, aqueous) . . . . . 1.00 gm.
- (b) Disodium hydrogen phosphate (anhydrous) . . . . . 5.00 gm.
- (c) Potassium dihydrogen phosphate (anhydrous) . . . 6.25 gm.
- (d) Distilled water . . . . . 500.00 c.c.

The phosphate salts are first dissolved in the water and then eosin added. It is set aside for 24 hours, filtered and is then ready for use.

### 4. TECHNIQUE OF STAINING

Blood films are ready for staining as soon as they are no longer obviously moist.

- a. Dip in solution A for 1-3 seconds.
- b. Rinse by gently waving in clear water for a few seconds, until stain ceases to flow from the slide, about 3-5 seconds.
- c. Dip in solution B for 1-3 seconds.
- d. Rinse in water for 3-5 seconds.
- e. Stand vertically on end to dry.

By standing the slide on end, the laked hemoglobin runs down and collects at one end of the film. Thus the background of the film will vary from colorless at the upper end to a deep pinkish-blue at the lower end where this hemoglobin has gathered.

The ultimate result of this stain is comparable to that achieved with Giemsa. The red of the chromatin is slightly brighter, but the cytoplasmic blue is not as deep or clear. The parasites retain their shape a little better than with the Giemsa method.

## 5. STAINING OF THIN FILMS

### a. Wright's stain

- (1) Cover the blood film with stock stain for  $1\frac{1}{2}$  minutes.
- (2) Add diluent in amount equal to the stock stain or until a metallic scum can be seen over the stain. Allow to stain for 3 minutes.
- (3) Wash in diluent until the film is pink in color.
- (4) Allow it to air dry.

### b. Giemsa stain

- (1) Fix blood film by immersion in absolute methyl alcohol for 1-2 minutes.
- (2) Allow to air dry.
- (3) Immerse in diluted stock stain, and stain for the desired length of time.
- (4) Wash in diluent for 1-2 minutes.
- (5) Allow to air dry.

TM 8-227 recommends a diluted stain of the ratio of 1 part stock Giemsa to 10 parts diluent, with a staining time of 20 to 30 minutes. There are many other dilutions that are advocated, such as:

1:15 for 15-30 min.  
 1:30 for 30-45 min.  
 1:50 for 45-90 min.

When a new stock stain is used, it is advisable to experiment with the various dilutions and staining times until the optimum differential coloration of the parasite is secured.

## 6. STAINING OF THICK FILMS

### a. Wright's stain

- (1) Dehemoglobinize the thick film by waving in water, 1/10 N. HCL, or 5% Formalin and 1% acetic acid mixture.
- (2) Wash thoroughly to remove the acid.
- (3) Allow to dry.
- (4) Stain as for a thin film.

### b. Giemsa stain

- (1) No fixing or dehemoglobinizing procedures.
- (2) Immerse in diluted stock stain for the desired staining time.
- (3) Wash in diluent for 1-3 minutes.
- (4) Allow to air dry. Do not blot.

In general, the higher dilutions (1:30 to 1:45) produce more complete dehemoglobinization in thick films and therefore produce a clear back ground against which the parasite may be more easily distinguished.

## 7. SOURCES OF ERROR IN DIAGNOSIS

Artefacts are numerous in thick films and are often confusing. Bacteria, dirt, and dust, precipitated stain, yeast cells and protozoa, molds, etc., are the chief source of these artefacts. Generally, they will be found to lie above the blood film plane, or may be refractile and focus unevenly. If clean slides and good technique are used in preparation of the film, there will be very few artefacts.

Platelets are often confused with parasites by the inexperienced technicians; especially, if they overlie a RBC. Practice alone will show the difference. This experience can be acquired in a relatively short time.

A good general rule to follow is to disregard anything that can be interpreted as an artefact; also, a diagnosis should never be made on the basis of one parasite, except under the most optimum conditions. For a definite diagnosis, the red chromatin and the blue cytoplasm, however distorted, must be recognized. Bacteria, spores, etc., may be mistaken for chromatin, so it is best to find several parasites before making definite commitment regarding a diagnosis; especially, a species diagnosis.

# SUMMARY OF PARASITE DIFFERENTIATION

Stained thin films			
	Plasmodium vivax	Plasmodium malariae	Plasmodium falciparum
Infected cell	Larger than normal, pale, often bizarre in shape. Schuffner's dots very often present. Multiple infection of erythrocyte not uncommon.	About normal or slightly smaller. Sometimes darker in early stages. Ziemann's dots rarely seen. Multiple infection of erythrocyte rare.	Normal in size, multiple infection of erythrocyte more frequent than in other species. Maurer's dots sometimes seen (in over-stained smears or when pH of H <sub>2</sub> O is on alkaline side.
			Plasmodium ovale
			Somewhat larger than normal, often with a fringed or irregular edge and oval in shape. Schuffner's dots appear even with younger stages, stain more readily and more deeply than in vivax.
Small trophozoite (early rings)	Signet ring form with heavy chromatin dot and large cytoplasmic circle, possibly with fine pseudopodia.	Signet ring form with heavy chromatin dot and cytoplasmic circle which is often smaller, thicker, and heavier than that of vivax.	Small, darker in color and more solid, as a rule, than those of falciparum. Schuffner's dots regularly present in almost 100% of infected cells.

# SUMMARY OF PARASITE DIFFERENTIATION

	Stained thin films		
	Plasmodium vivax	Plasmodium malariae	Plasmodium falciparum
Growing trophozoite	<p>Same as above with gradual increase in amount of cytoplasm and chromatin. Often with more distinct pseudopodial processes. Small yellowish-brown pigment granules in cytoplasm, number increasing with age of parasite.</p>	<p>Chromatin rounded or elongated cytoplasm in a compact form with little or no vacuole or in a narrow band form across the cell. Dark brown pigment granules may have peripheral arrangements.</p>	<p>This stage remains in the ring form but chromatin and cytoplasm increase to the extent that in size the parasite resembles closely the small trophozoite of vivax. A few pigment granules give a yellowish tinge to the cytoplasm. This is usually the oldest asexual stage seen in peripheral circulation</p>
Large trophozoite	<p>One abundant mass of chromatin, loose, irregular or close compact cytoplasm, with increasing amounts of fine brown pigment. Parasite practically fills enlarged cell by end of 36 to 40 hours.</p>	<p>One mass of chromatin, often elongated, frequently less definite in outline than that of vivax. Cytoplasm dense, compact with few irregularities of outline; in rounded, oblong or sometimes band shape. Pigment granules larger, darker than in vivax with great tendency toward peripheral arrangements. Fills or almost fills normal cell.</p>	<p>Stage seldom seen in peripheral blood. Very small, solid, with one small mass of chromatin; lightly staining, compact cytoplasm; and with a haze of pigment scattered through the cytoplasm or with very dark pigment collected in one small, dense block.</p>
			<p>Resembles closely same stage of P. malariae but is considerably larger. Pigment is lighter in color and less conspicuous.</p>

# SUMMARY OF PARASITE DIFFERENTIATION

Stained thin films				
	Plasmodium vivax	Plasmodium malariae	Plasmodium falciparum	Plasmodium ovale
Schizont (presegmenting)	Chromatin divided into a number of masses, cytoplasm shows varying degrees of separation into strands and particles; pigments show tendency to collect in parts of the parasite.	Same as vivax except that the parasite is smaller and shows fewer divisions of chromatin, as it approaches segmentation, and more delayed clumping of pigment.	When found in peripheral blood, this stage resembles the same stage of P. malariae, but is smaller and the pigment is likely to be completely clumped in one small dark mass.	About 25% of infected cells are definitely of oval shape. Usual picture is that of a round parasite in center of an oval cell. Many cells with indefinite fringed outline. Pigment lighter in color and less coarse than in P. malariae.
Mature (Schizont)	12 to 24 divisions or merozoites, composed of a dot of chromatin and a portion of cytoplasm. The pigment is in one or two clumps. Parasite practically fills enlarged cell.	6 to 12, usually 8 or 10, merozoites in a rosette or irregular cluster. Practically fills normal sized cell.	8 to 24 merozoites, which are very small compared to those of other species. Rarely found in peripheral blood. Fills about two-thirds of normal sized cell.	Usually 8 merozoites arranged around a central block of pigment.

## Stained thin films

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# SUMMARY OF PARASITE DIFFERENTIATION

	Stained thin films			
	Plasmodium vivax	Plasmodium malariae	Plasmodium falciparum	Plasmodium ovale
Length of asexual cycle.	48 hours	72 hours	48 hours	48 hours
Stages in peripheral blood.	All	All	Usually ring form trophozoites and gametocytes. Other stages rarely found except in severe cases.	All
Remarks	More stages of growth likely to be seen in one film than in other species. Gametocytes appear early in cycle.	Parasites are usually more compact and hence appear more intensely stained than those of other common species. Gametocytes rarer than in other common species, appear late. Least often found of 3 species in United States.	Parasites frequently more numerous than in other infections. Unlike other species growth of asexual forms, following the ring stage, takes place in internal organs.	Species not found to date in United States. Differentiation not possible in thick films.

FROM N.I.H. BULLETIN #180, U.S.P.H.,  
AIMEE WILCOX, pages 16-17.

ENTOMOLOGICAL PROCEDURES



## MOSQUITO DISSECTION

### 1. DISSECTION OF THE MALE TERMINALIA

The last two of three segments are clipped from the tip of the abdomen of a male mosquito. This operation should be performed with a very small sharp pointed pair of scissors.

The male terminalia is then placed in a small crucible (with a handle) which is filled about one-half full with a 10% KOH solution. The crucible is held over a flame, and is removed as soon as steam appears. By means of a small pipette, the male terminalia is transferred from the KOH solution to a clean glass slide. The excess KOH is blotted off with pieces of filter paper and the terminalia covered with a small drop of Gater's solution. The slide is then focused under a dissecting microscope with 10X oculars and 4X objectives. By the use of small minuten needles set in wooden handles, the male terminalia is manipulated and dissected until all parts come in view. These should be placed on one end of the slide so that a label showing the location, specimen number, date and species can be affixed on the other end. A cover slip is placed over the drop containing the male terminalia which can be studied more closely if necessary. Mounts should be stored in a slide box for further reference.

### 2. GATER'S FORMULA

Gum Arabic (Picked) . . . . .	grams	8
Distilled water . . . . .	millilitors	10
Chloral hydrate . . . . .	grams	74
Glucose syrup (10 grams of glucose in 10 mls. of water)	millilitors	5
Glacia acetic acid . . . . .	drops	3

### 3. DISSECTION OF STOMACH AND GLANDS (From Human Malaria, page 54 to 58)

#### a. STOMACH

Engorged wild mosquitoes should be kept in a cool incubator for several days prior to dissection, until, through digestive processes, the stomach is emptied. Kill not more than two or three mosquitoes at a time with chloroform. If several are killed at once, place those not immediately required in a moist chamber. After identification, cut off the wings and legs. Keep these in a properly labelled pill box for future reference. Place the mosquito on a slide on the stage of a dissecting microscope with the abdomen pointing into a drop of saline. Secure the mosquito on its back or side by piercing the thorax with the point of a straight needle held in the left hand. With a curved needle, held in the right hand, nick the seventh or eighth segment above and below. Then insert the needle point in the last segment, and pull gently, drawing out the intestinal tract and appendages into the saline solution. Trim off the malpighian

tubes, leaving enough to identify the posterior end, and cut through the fore and hind guts close to the stomach. Transfer the stomach, on the needle tip, to a clean drop of saline. Straighten it out and cover with a cover slip for examination in fresh condition. If excessively contracted, the stomach may be expanded by cautiously warming the slide.

In the dissection of gravid mosquitoes, it may be difficult to remove the stomach in the manner described since the ovaries block the opening and the tissues may break. In this case, tear the membranous sides of the abdomen with the needle point and dissect out the stomach.

#### b. SALIVARY GLANDS

Various techniques have been described, some of which may be better adapted to certain technicians than others. Practice is more important than method. Place remainder of the mosquito body in a drop of saline tinted with methylene blue. With the head and thorax pointing to the right, hold the mosquito in place with the needle in the left hand. Cut off the head with the curved needle held in the right hand. Press the thorax gently with the side of the curved needle, forcing a small mass of tissue from the neck opening. The tips of the glands may be seen protruding from this mass and can be gently teased out. If they are not seen, tease the greenish-colored mass apart in search of the two glistening, blue-stained bodies. Care should be exercised to assure that the glands have not been drawn from the thorax as the head is removed from the thorax. Transfer the glands, free of other tissues, by the curved needle tip to a clear drop of saline and cover with a cover slip. (During the process of transferring, care must be exercised not to lose the glands on the tip of the needle. - Note by Colonel John E. Elmendorf, Jr.).

#### 4. EXAMINATION OF FRESH PREPARATIONS

The preparations are examined with a compound microscope equipped with a mechanical stage.

##### a. STOMACH

Focus a two-thirds objective on the stomach and move it to center of the field. Place the tip of a needle held in the left hand against the edge of the cover slip. Move the slide by means of the mechanical stage against the needle point, so that the cover slip is held fixed. This causes the stomach to roll, so that all parts of its surface may be examined for projecting cysts. If suspicious bodies are found, they are examined under a higher magnification.

Small cysts are seen as clear, round, or oval bodies without a refractile wall, but with unmistakable pigment granules. Large cysts show a distinct wall and only rarely is pigment visible. Mature cysts are finely striated, owing to the myriads of sporozoites which they contain. When only a few cysts are found, they are usually in the posterior end of the

stomach.

The contraction of some of the circular muscle fibers in the central part of the stomach may produce externally extruding sacs of stomach membrane that may cause confusion. Some of the fat cells containing fat globules might also be confused with cysts. Small cysts should never be diagnosed unless pigment is seen. Large cysts are unmistakable.

#### b. SALIVARY GLANDS

Move the glands to the center of the field of a two-thirds objective. With the tip of a needle press gently on the cover slip above so as to crush them. Then the fluid about the crushed glands is examined under a one-sixth objective for sporozoites. These are slender, glistening rods, straight or slightly curved, and about 12 to 14 micra in length. Motility, at least in the sense of a translation of position, is questionable.

## STAINING OF PREPARATIONS

### 1. STOMACH

If a high incidence of infection in the mosquitoes is expected, it is preferable to fix and stain the stomachs before examination. Fixation and staining of specimens which have been for some time under a cover slip in saline is unsatisfactory. By staining freshly dissected stomachs in Mayer's acid haemalum (formula follows) before examination, infections may be found which would be missed if examination is limited to the fresh preparation. Cysts, while small, are detectable as early as the third day after an infective feeding in stained specimens, while such small cysts are very difficult to detect in unstained preparations.

After a stomach is removed and Malpighian tubes trimmed off, it is covered with a large drop of Bouin's Fixative. Then the fore gut and hind gut are cut off. Allow the fixative to act for five minutes. The specimen is transferred with a pipette throughout the different steps of the staining process. After fixation, the preparation is rinsed in distilled water until all traces of the yellow color of the picric acid have disappeared. This requires several hours or preferably over night. From the distilled water the stomach is transferred to a 1:10 dilution of Mayer's acid haemalum for an hour. The stomach is next washed to remove excess stain in a one percent solution of acetic acid for three to five minutes, then transferred to a one percent solution of sodium carbonate to neutralize. (The color changes to blue). For dehydration, it is passed successively through 50, 70, 85, 95, and 99 percent solutions of alcohol, being allowed to remain for three to five minutes in each. It is then cleared in carbol-xylol and mounted in balsam underneath a cover slip.

Formulae for the stains mentioned are as follows:

#### a. MAYER'S ACID HAEMALUM

Dissolve 1 gram of haematin in 50 c.c. of 90 percent alcohol and warm. Add this solution to a solution of 50 grams of aluminum potassium sulphate in 1000 c.c. of distilled water dissolved by heating. Mix warm, cool, and filter. Add 2 c.c. of glacial acetic acid to 100 c.c. of haemalum solution.

#### b. BOUIN'S FIXATIVE

Picric acid, saturated aqueous solution . . . . .	30 parts
Formalin (40 percent) . . . . .	10 parts
Acetic acid . . . . .	2 parts

This should be prepared daily.

### 2. SALIVARY GLANDS

After the preliminary examination, the cover slip is gently raised

from the crushed gland and inverted. The material on both the side and the cover slip is allowed to dry, is then fixed by flooding with absolute methyl alcohol, and is stained with Giemsa stain exactly as in the case of blood smears. For staining, forceps for holding the cover slips are very convenient. The surface of both the slide and cover slip is examined with a low power lens and the one to which the glands have adhered is saved. If they are on the cover slip, the slip is attached, gland side upward, to a slide by a drop of balsam. (Boyd 1932).

### 3. EXAMINATION OF STAINED PREPARATIONS

A well stained stomach preparation shows the cysts slightly darker than the stomach tissue, with the pigment showing as in a fresh preparation. Large or mature cysts are readily recognized by their internal content of sporoblasts or sporozoites. Small cysts are identifiable by their content of pigment, the presence of which is indispensable for diagnosis of the body as a cyst. The only criterion for the specific identification of the cysts is the characteristic appearance of the pigment, which is retained from the macrogametocytes from which they developed. Pigment is rarely observed in the large cysts. It is not certain whether its disappearance is apparent or real, i.e., whether it is masked by the increased internal structure of the cyst or has been metabolized. The stained sporozoites exhibit the characteristic colors of a malarial parasite. The reddish chromatin is usually a central oval mass or may appear as three or four small, round granules. The cytoplasm has the familiar blue of the stained parasite. The sporozoites of P. vivax and P. falciparum are morphologically indistinguishable, but one could probably differentiate the sporozoites of P. malariae, as these are definitely larger and coarser, with splotchy chromatin. Sporozoites from different mosquitoes vary greatly in their size, although those derived from any particular insect show relatively slight difference. (Boyd 1935a).

## ORGANIZATION AND PROCEDURES FOR SAMPLING FAUNA

Maps should be secured or, if necessary, prepared on which are shown the location of breeding areas, major land marks, highways, roads, railroads, and streams. The location of catching stations (natural resting places), light traps, etc., should be plotted on the map. (See Figure 1). For entomological work a map made to a scale of 1 inch to 660 feet is preferable.

### 1. LOCATION OF BREEDING AREAS

All known breeding areas, discovered and proven through exhaustive field studies, should be located on maps. New areas should be added to the maps as they are located. It is a prerequisite for orientation of work of control that each breeding area be identified by means of a numbering device.

A separate list should be made of the breeding areas, showing the number, type of breeding area, and distance from locality to be protected.

Every conceivable type of breeding place must be taken into consideration, such as wells, cisterns, barrels, tubs, tree holes, air plants, ponds, lakes, marshes, salt marshes, seepage areas, streams, rivers, gulfs, and marginal areas of oceans.

### 2. LOCATION OF ADULT CAPTURE STATIONS

A person with entomological training should investigate the area and select the types of places where adult anophelines are known to congregate. The greatest percentage of anophelines will be found resting in locations which are humid, dark, and protected from wind currents, such as culverts, under houses, in animal stables, in houses, hollow trees, caves, etc.

It will be found that certain anophelines have peculiar resting habits, and do not rest in the above type of places. Some species rest in vegetation over the water surface, etc. Some species will enter huts and sleeping quarters to feed, but will not remain inside during the day; others will remain inside for considerable periods.

The stations selected should form a meshwork over the entire area that is being investigated, and also be so located that they serve as a coverage for adequate sampling of known breeding areas, and as a measure of areas to be protected. It is necessary that each station be numbered. A list should be made showing the station's number, type and distance from area to be protected.

### 3. LOCATION OF LIGHT TRAPS

In the location of light traps, it is advisable that two precautions

be observed: Situate the trap about six feet above the ground in such position that it will be protected from other interfering lights, which may cause a diffusion of attraction, and ascertain that the trap is duly protected from disturbing wind currents. Each of the light traps employed must be designated on an area map with a significant number of identification purposes.

#### 4. LOCATION OF ARTIFICIAL CONTAINERS AS RESTING PLACES

When barrels, boxes, etc., are used as natural resting places, they should be numbered. It is preferable to locate these containers in dense clumps of bushes, shrubs and under trees, and preferably where it is shaded and damp.

#### 5. LOCATION OF ANIMAL BAIT TRAPS

A portable bait trap of the Magoon type may be placed on a trailer, or a trailer might be made into an animal bait trap that could be transported from place to place as desired.

## METHODS OF CAPTURING ADULT ANOPHELINES

### 1. CAPTURES FROM NATURAL RESTING PLACES

#### a. TYPES OF PLACES INDICATED FOR SEARCH FOR RESTING ADULT MOSQUITOES

- (1) In houses
- (2) Under houses
- (3) In barns
- (4) In animal stables
- (5) Hollow trees
- (6) Out-houses
- (7) Vegetation
- (8) Latrines
- (9) Caves
- (10) Stump holes
- (11) Under culverts and bridges

#### b. METHODS OF CAPTURING MOSQUITOES FROM NATURAL RESTING PLACES

- (1) Captures made by means of chloroform tube. (See Figure 2).
- (2) Preparation of chloroform tube, and types of tubes that may be utilized.
  - (a) Large test tube.
  - (b) Alka-Seltzer bottle.
  - (c) Plastic bottle 1" x  $4\frac{1}{2}$ ". (Can be purchased from:  
Ward's Natural Science Establishment, Inc.,  
Rochester, New York).

Bottom of the tube is filled with cut rubber chips to a depth of about one inch. Two or three pieces of soft cardboard are cut in circular form to fit inside the tube and these are placed over the rubber. Chloroform is poured into the tube until the rubber is saturated. Frequently shreds of cut, soft paper are inserted into the tube above this mixture to prevent joggling of mosquitoes while in tube and resulting ruination of their markings.

- (3) Captures made by means of long glass tube. (See Figure 3).

The size of this glass tubing should be from one to two feet long and  $5/8$ " in diameter. A piece of doubled bobbinet should be placed over the end of the tube. Cotton may also be placed in the tube to prevent passage of mosquitoes into the rubber tubing. A rubber hose is then adjusted over the bobbinet or the end containing the cotton.

Mosquitoes captured by this method of suction by the mouth should be blown into another container. The type of containers used for this purpose will naturally depend on the type available in the locality. Types that are most commonly used are: Lantern chimneys, wide-mouth bottles, and cages that can be constructed from wood and steel frames, as well as pasteboard boxes. Where pasteboard boxes are used, the sides of the pasteboard boxes are cut out. Bobbinet is then stretched over the box frame. A sleeve can be adapted to this box so that the glass tube containing the mosquitoes may be introduced into the sleeve and the captured mosquitoes blown into the cage. Rubber bathing caps, with two slits cut at right angle to each other, may be placed over lantern chimneys, or the mouth of bottles. The glass tube containing the mosquitoes may then be introduced into the receptacle through the slits in the bathing cap, and the mosquitoes blown into the cage.

(4) Captures by means of specially constructed catching tubes.

Commercial types of capture tubes may be purchased which are made of glass or plastic, and are approximately  $1\frac{1}{2}$  inches in diameter by 6 inches long. Plastic toothbrush holders may be constructed into a catching tube by cutting off the bottom rounded end of such a plastic container. A small hole is made at the apex of the cone, and this funnel then inverted and sealed in place at one opening of the tube. A cork or rubber stopper is placed on the opposite end. Through a hole in the cork or stopper, a short glass tube is inserted which fits flush on one side of the cork and is allowed to protrude about one inch on the outside of the cork. A small and suitable diameter rubber tube is adapted to the protruding end of the glass tube. Before sucking the mosquitoes into the tube, it is advisable to place loosely packed cotton inside the tube at the stopper or cork end in order to prevent sucking mosquitoes into the mouth. The cotton in the tube also facilitates killing the mosquitoes later. A few drops of chloroform dropped on the cotton when the stopper is extracted will kill the mosquitoes in a few seconds and they can be then taken from the tube and placed in a pill box, properly labeled. (See Figure 4).

2. HAND CAPTURES WITH HUMANS OR ANIMALS USED AS BAIT

Captures by means of human or animal bait should be made at the period when mosquitoes are known to feed. This generally is from dusk on into the night. When night collections are made, a flashlight and catching tube are essential.

Individuals who are collecting mosquitoes from each other's person, or

from animal bait, should by all means wear head nets, gloves and sufficient clothing to insure that mosquitoes will not be able to penetrate and to bite. The individual who is working in a malarious district and does not take these precautions will, in all probability, contract malaria.

### 3. CAPTURES FROM ANIMAL BAIT TRAPS

An animal bait trap is a type of enclosure that will confine an animal, and so constructed that mosquitoes can enter, but once inside, will encounter difficulty of exit.

There is one type of animal bait trap devised by Magoon (1) that is constructed in the form of a small house about four feet wide, by eight feet long, by seven feet high, with a small one inch opening or slot around the center of the cage, midway between the ground and roof, through which slot the mosquitoes can enter. The bottom section of the trap, from the ground up to the entrance slot, is made of tin, galvanized metal, or lumber. Above this lower section and all around the trap, the above mentioned slot for entrance of mosquitoes is located. This slot is made by using pairs of boards about four inches wide by one inch thick, and cut to the appropriate lengths corresponding to the length and width of the trap. These are brought together in a V shape throughout their entire length, leaving an opening at the apex of the V which is pointed inside the cage and which is about one inch wide. The top half of the cage is covered with screen wire. The roof may be made of tin or wood.

An animal (cow, horse, or donkey), according to the blood meal preference of the anopheline vector in the locality, is confined within the cage at night. In the morning the animal is taken out. The collector enters the cage and collects the mosquitoes by means of a catching tube.

In South America, Shannon (2) constructed an animal bait trap of mosquito netting which was suspended to trees and covered a horse, allowing, of course, openings for mosquitoes to enter.

### 4. CAPTURES FROM LIGHT TRAPS

A type of light trap that has been most successfully used is the New Jersey Light Trap. (See Figure 6). The attraction of mosquitoes to light varies with species. Some species are attracted readily while others react to a lesser degree.

The principle on which the light trap functions is that mosquitoes are attracted to the light in the top of the trap. Above the cylinder and below the light is suspended an electric fan, which, by means of suction, draws the mosquitoes from the light, blowing them to the bottom of the cylinder through a screen cone into a cyanide killing bottle. (See Figure 5). The efficiency of the light trap has not been demonstrated for all species of anophelines. Negative findings for the presence of anophelines should be accepted only in conjunction with other investigative procedures. For

example, in our hands, the light trap does not function well with A. quadrimaculatus in Florida.

5. CAPTURES FROM BOXES, BARRELS, ETC.

It has been found by placing boxes, barrels, and kegs, painted black or red on the inside, in clumps of bushes, under trees and in dense vegetation, that such artificial containers make fairly satisfactory daytime resting places for certain species of Anopheles. Smith (3) used kegs placed at strategic points around an impounded lake in Tennessee. Russell and Santiago (4) used an earthlined trap as a daytime resting place for anophelines in the Philippine Islands.

## METHODS OF HANDLING ADULT MOSQUITOES LIVING AND DEAD

### 1. LIVING SPECIMENS

Adult mosquitoes which are to be kept alive for experimental purposes should be placed in lantern globes, small cages, or wide mouth jars. The mosquitoes must then be stored in a cool humid place. They should be fed on a 5% sugar-water solution. If subsequent batches of ova are required from the female adults, it will be necessary to allow them to feed on rabbits or on other available animals. The live mosquitoes must always be protected from ants and other insects. This can be accomplished by placing table legs on which specimens are placed in pans of oil. Ant barricades can be made by placing cloth bands saturated in a solution of bi-chloride of mercury and alcohol. Allow these bands to dry and then wrap the bands around table legs, light wires, etc.

Live mosquitoes which are to be shipped short distances should be placed in small cages, (6" x 6"), made of screen wire or bobbinet. It will be necessary to place a band of cotton about one inch wide, saturated with 5% sugar water, around the cage. The cage should be wrapped in wax paper and placed in a sturdy box (wood or cardboard) for shipment.

Mosquitoes that are to be transported for any great distance, or when the temperature is in excess of 85° F., should be carried in small ice chests. The chest most commonly used is the small metal automobile ice box.

Ova may be obtained from wild caught females by placing a female mosquito in a shell vial containing a piece of moistened filter paper placed to fit about one half of the inside surface of the side of the vial. The vial should then be placed on its side with a moistened filter paper at the bottom. The ova can be collected from the filter paper with a wire loop or they can be washed from the filter paper with a pipette.

When a great number of ova are required, it will be desirable to enclose ten or fifteen female mosquitoes in a lantern chimney or small cage made from screen wire or bobbinet. The chimney or cage should be placed over a cup or one or two small strips of cork on the water surface so that the females may have a resting place for the function of oviposition, as well as to prevent their being trapped in the surface film of the water.

Ova can be taken from the surface of the water by means of a spoon or wire loop and placed inside a cork or waxed paper ring which is floated in a receptacle holding water.

### 2. DEAD SPECIMENS

#### a. PROCEDURES OF HANDLING ADULTS IN THE FIELD

Mosquitoes captured from stations and animal bait traps, and not for

use as live specimens, should be killed with chloroform. The dead mosquitoes are placed in labeled pill boxes showing: (1) Locality, (2) Station Number, and (3) Date, (4) Collector's Name, (5) Type of Shelter (Trap, etc.).

#### b. SHIPMENT OF MOSQUITOES

Mosquitoes that are to be shipped in pill boxes should have label and locality, date, collector, whether reared or captures, (Type of trap, etc.). A small piece of crumpled tissue paper should be placed in the bottom of the box, together with melted naphthaline (crushed moth balls), and a piece of crumpled tissue should be placed lightly over the mosquitoes as well. This is done in order to prevent the mosquitoes from being mutilated by excessive shifting inside the pill boxes. A soft type facial tissue paper will be found to be superior to most types of cotton. Mosquitoes packed in ordinary cotton frequently become entangled in the cotton, and upon removal are found ruined for identification purposes.

#### c. MOUNTING MOSQUITOES

Mosquitoes may be mounted on minuten nadeln pins, when available, on cork blocks, or cardboard points, the latter having a small drop of cellulose cement placed on the point of the cardboard before touching it to the mosquito. (See Figure 7).

#### d. STORAGE OF MOSQUITO SPECIMENS

Mosquito specimens may be stored in Schmidt boxes, or in individual shell vials. It is necessary to keep paradichlorobenzene or naphthalene crystals in the storage boxes as well as in the individual vials. If mosquitoes are to be stored in pill boxes, it is desirable to place some crystals of paradichlorobenzene or naphthalene under the tissue placed on the bottom of the box. The box may be sealed by placing scotch tape over the seam between the cover and the bottom of the box.

## METHODS OF COLLECTING LARVAE FROM BREEDING AREAS

### 1. COLLECTION OF LARVAE FROM PONDS, MARSHES, STREAMS, ETC.

In the great percentage of breeding areas the larvae are generally found in the vegetation, around logs or debris. In a clear small pool, the larvae will be found circling the edge of the pool. As a special procedure applicable to small puddles or depressions where larvae are not readily demonstrable, they can, at times, be made to rise to the surface by mudding the pool. Some species of anopheline larvae, when disturbed, will go to the bottom of the breeding area, and will remain on the bottom for some time. Care should be taken not to disturb the surface film before dipping. If an individual should wade into a pond and not find anopheline larvae after dipping, it will be found advantageous to remain motionless for a few minutes to give the larvae an opportunity to rise to the surface before proceeding to dip.

#### a. COLLECTIONS OF LARVAE WITH DIPPERS

A small white enamel dipper with an attached handle about four feet long is probably the most satisfactory type of equipment with which to collect larvae. The ability to recognize favored location for presence of larvae and, accordingly, those spots most favorable for dipping develops with practice. The most propitious places for discovery of larvae are those where horizontal and vertical vegetation are present. A "dip", better termed a "sweep", is made by skimming under the surface film. If anopheline larvae are present, they are most frequently seen lying parallel to the surface and just under the surface film of the water in the dipper. The larvae can be transferred from the dipper to a vial or jar by means of a wide mouth pipette or medicine dropper. (See Figure 10).

#### b. LABELING THE VIALS OR JARS CONTAINING SPECIMENS FROM THE BREEDING AREAS

The larvae are placed in vials or jars. (See Figure 11). The receptacles are labeled, indicating locality, number of breeding area, and date of collection.

#### c. PREPARATION OF LARVAE FOR SHIPMENT TO A CENTRAL LABORATORY

In the field, the larvae are put into vials by means of a wide mouth pipette or medicine dropper. In so doing, a considerable amount of water from the breeding area is likewise introduced into the vials. All excess water should be siphoned out. The vials can then be filled to the top with a 10% formalin solution or 70% alcohol. It is advisable to expel all air bubbles from the vial. This can be accomplished by inserting a pin between the cork and inside of the vials, while pressing on the cork. As a general rule, it is not necessary to kill the larvae before putting in the formalin or alcohol as these agents will themselves accomplish this end. Larvae

may likewise be killed by placing specimens in water at a temperature of 120° to 140° F. The larvae should be killed in hot water when permanent slide preparations are desired. The larvae can be transferred from the hot water to Gater's solution, with a pipette if desired for permanent specimens.

#### d. CARE OF LARVAE TO BE BRED FOR IDENTIFICATION

The larvae that are to be bred, such as the small larvae, should be brought into the laboratory in vials or jars, and transferred to pans that are labeled with the information specifying location of collection, number of breeding area, and date.

If living larvae are to be transported great distances, it is occasionally essential to transport them in ice chests.

#### e. TECHNIQUE OF REARING LARVAE

A number of techniques have been described for rearing anopheline larvae in the laboratory. Boyd, et al, (5) described one of the early techniques that consisted of feeding larvae on ripened hay infusions supplemented with Fleishmann's Yeast.

Other types of feeding methods have been described, such as feeding larvae on wheat infusions, powdered dog biscuits, powdered "Pablum", powdered blood, Brewer's Yeast, killed algae, etc. Bates (6) has described a method of rearing larvae by filling pans full of water from the breeding areas themselves and placing, as well, some mud from the breeding area in the bottom of the pan. A few stale bread crumbs are added to the pans at frequent intervals.

### 2. COLLECTION OF LARVAE FROM AIR PLANTS

The collection of larvae from air plants can best be accomplished by removing the plants from the trees. The plant should be held in an upright position while it is being removed from the tree so that the water collected at the leaf bases will not be spilled. The leaves are carefully cut off and the plant immersed in a bucket of water. They should be left for several hours and as the larvae are separated from the plant, they can be moved into another receptacle.

### 3. COLLECTION OF LARVAE FROM TREE HOLES

Larvae can be collected from tree holes by using a pipette made by placing a large rubber bulb over a hollow glass tube. (See Figure 9). The water in a tree hole can be siphoned out and placed in a suitable receptacle. It is also possible to take the leaf mold from tree holes that are dry and place in boxes or tin cans for transportation. The mold is then placed in pans and flooded with water. If any ova are present in the leaf mold, larvae should develop later in the pans.

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1. Magoon, E.H., 1935. A Portable Stable Trap for Capturing Mosquitoes. Bull. Ent. Res., 26:363.
2. Shannon, R.C., 1939. Methods for Collection and Feeding Mosquitoes in Jungle Yellow Fever Studies. Amer. Jour. Trop. Med., 19:131.
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4. Russell, P. F. and Santiago, D., 1934. An Earthlined Trap for Anopheline Mosquitoes, Proc. Ent. Soc., Wash., 36:1.
5. Boyd, Mark F., Cain, T.L., Jr., and Mulrennan, J. A., 1935. The Insectary Rearing of Anopheles quadrimaculatus, Amer. Jour. Trop. Med., 15:285-402.
6. Bates, Marston, 1941. Studies in the Technique of Raising Anopheline Larvae. Amer. Jour. Trop. Med., Vol. 21, No. 1, 103-122.

# DETAIL OF ENTOMOLOGICAL MATERIALS AND EQUIPMENT FOR MALARIA INVESTIGATION

WITH ESTIMATE OF QUANTITIES FOR ONE UNIT OF ONE OR TWO ENTOMOLOGISTS

## 1. SMALL EQUIPMENT

- |  |                                 |
|--|---------------------------------|
| (1) 2 two-celled flashlights                                     | (29) 4 strips of sheet cork,    |
| (2) 24 batteries and 6 bulbs                                     | 4" x 8" x $\frac{1}{2}$ " White |
| (3) 2 dippers (white enamel)                                     | paper slides.                   |
| (4) 24 pipettes (12 wide mouth, 12 small)                        | (30) 4 dissecting needles,      |
| (5) 1 lb. glass tubing $\frac{1}{8}$ " and $\frac{1}{2}$ " diam. | (2 straight, 2 curved)          |
| (6) 24 plastic killing tubes, 1" x $\frac{1}{4}$ "               | (31) 100 aluminum blanks        |
| (7) 6 ft. rubber tubing to fit $\frac{1}{8}$ "                   | 1" x 3" x 1.5 mm.               |
| (8) 2 gross small, tin pill boxes                                | (32) 2 small scissors           |
| (9) 1 gross large, rectangular pill boxes                        | (33) 4 forceps (2 curved,       |
| (10) 1000 shell vials with corks                                 | 2 straight)                     |
| (11) 2 lbs. cotton   | (34) 2 scalpels                 |
| (12) 2 yds. of white & black bobbinet                            | (35) 5 notebooks                |
| (13) 4 rubber bulbs (large)                                      | (36) 2 bath tub thermometers    |
| (14) 5 dropping bottles  | (37) 2 hunting knives           |
| (15) 1 box facial tissue   | (38) 2 cartridge belts          |
| (16) 12 small cream pans   | (39) 2 machettes                |
| (17) 10 hollow ground slides                                     | (40) 2 snake bite kits          |
| (18) 2 crucibles with handles                                    | (41) 1 box filter paper         |
| (19) 24 test tubes   | (42) 2 carrying cases,          |
| (20) 12 wide mouth pint bottles with                             | shoulder straps                 |
| stoppers (rubber)  | (43) 280 glass slides           |
| (21) 2 pairs of leather gloves                                   | (44) 6 boxes labels, assort-    |
| (22) 2 black head nets   | ed sizes                        |
| (23) 2 hand lens   | (45) 4 boxes glass cover        |
| (24) 1500 insect minuten pins                                    | slips                           |
| (25) 500 insect minuten pins                                     | (46) 4 slide boxes, 100         |
| (26) 500 insect cardboard points                                 | box capacity                    |
| (27) 4 strips of sheet cork,                                     | (47) 2 deep well slides         |
| 4" x 8" x $\frac{1}{4}$ "  | (48) Graduates 2-10 c.c.,       |
| (28) 6 white enamel cups   | 2-50 c.c., and 2-1,000          |
|  | c.c.                            |

## 2. LARGE EQUIPMENT

- |                             |                          |
|-----------------------------|--------------------------|
| (1) 1 dissecting microscope | (4) 1 hand axe           |
| (2) 1 compound microscope   | (5) 4 New Jersey Light   |
| (3) 2 microscope lamps      | traps                    |
|                             | (6) 1 automobile ice box |

### 3. CHEMICALS AND MISCELLANEOUS

- |  |   |
|--|---|
| (1) 4 pts. chloroform                      | (20) 1 pt. xylene                         |
| (2) 4 pts. formaldehyde                    | (21) 2 qts. ethyl alcohol                 |
| (3) 1 pt. pear oil                         | (22) 4 oz. Bottle methylene blue          |
| (4) 1 lb. polyvinyl alcohol                | (23) 100 grms aluminum potassium sulphate |
| (5) 1 lb. potassium hydroxide              | (24) 4 oz. bottle Giemsa stain            |
| (6) 4 tubes cellulose cement or shellac    | (25) 4 oz. balsam                         |
| (7) 1 pt. phenol                           | (26) 1 lb. sodium hydroxide               |
| (8) 1 pt. lactic acid                      | (27) 5 gr. acid fuchsin                   |
| (9) 10 gr. haematin                        | (28) 1 pt. con. HCL                       |
| (10) 4 oz. picric acid                     | (29) 1 pt. acetone                        |
| (11) 8 oz. sodium chloride                 | (30) 4 oz. clove oil                      |
| (12) 1 pt. methyl alcohol                  | (31) 8 oz. modeling clay (plasticine)     |
| (13) 1 pt. glacial acetic acid             | (32) 1 diamond point pencil               |
| (14) $\frac{1}{2}$ lb. gum arabic (picked) | (33) 1 pk. colored pencils                |
| (15) 1 lb. chloral hydrate                 | (34) 2 pk. lead pencils                   |
| (16) 1 qt. glycerine                       | (35) 2 oz. cedar oil                      |
| (17) 2 lbs. sodium cyanide crystals        | (36) 8 oz. crown immersion oil            |
| (18) 2 lbs. paradichlorobenzene            |   |
| (19) 1 lb. dextrose                        |   |

### MOUNTING PREPARATIONS

#### 1. FORMULAS USED FOR MOUNTING LARVAE, ADULTS, AND MALE TERMINALIA

##### a. Gater's solution modified

Gum arabic (picked)	8 grams
Distilled water	10 c.c.
Chloral hydrate	74 grams
Glycerine	5 c.c.
Glacial acetic acid	3 c.c.

##### b. Polyvinyl alcohol: A medium for mounting and clearing biological specimens.

Polyvinyl alcohol	
(Stock solution, consistency of syrup)	60%
Phenol	20%
Lactic acid	20%

#### 2. SOLUTIONS USED FOR PRESERVING OVA, LARVAE, AND PUPAE

##### a. 10% formalin

##### b. 70% alcohol

##### c. MacGregor solution:

50% solution formalin	10.00 ml.
5% solution borox	10.00 ml.
Distilled water	80.00 ml.
Glycerine	0.50 ml.

## GENERAL ESTIMATE OF EQUIPMENT FOR MALARIA INVESTIGATION AND CONTROL

RECOMMENDED MINIMUM EQUIPMENT FOR EACH UNIT, TO BE SENT WITH UNITS. ---  
(For Complete Entomological Equipment, see page 147).

### 1. Larvicidal Equipment

Knapsack or cylindrical can sprayers or their equivalent. (For use with oil and Paris green water mixtures, Barber methods).

Paris green dusters.

Screen wire for making Paris green mixtures. (Approximately 38 mesh to the inch).

Paris green.

DDT powder.

Oil, if not securable in locality.

Boots and long heavy-duty gloves.

### 2. Engineering Equipment

Line or string level.

Malaria survey map-making equipment:

Tripod (can be made locally from bamboo or wooden poles tied with leather thong.

Plane table (most likely can be secured locally in almost all localities).

Watch compass.

Triangular rule - Engineer's scale.

Small straight-edge and triangle (45-45, optional 60-30).

50 foot tape (optional).

Hulse's Instructions for Field Mapping, or its equivalent.

### 3. Clinical Investigative Equipment

Glass slides for blood smears, enough for entomological uses as well.

Stains for malaria parasites.

Compound microscope.

Needles for securing blood drop for malaria slides.

Slide boxes (one to be used as sample for local manufacture).

Lens paper.

Alcohol.

Cotton.

Colored pencils for marking on glass slides.

Immersion oil.

Xilol.

4. Equipment for Disease Prevention and Cure

Medications: Quinine, Atabrine, etc.

Individual cot mosquito nets with tapes for tying in place located at four corners.

Repellents.

Spray guns for killing adults (or pyrethrum bombs).

Pyrethrum extract or Westinghouse aerosol bombs.

Head nets.

5. Entomological Equipment

Small pocket magnifying glass (10 power).

Mosquito capture tubes.

Lantern chimneys or their equivalent for keeping adults.

Larval collection dippers.

Small glass vials with stoppers for larval transportation after collections.

Large mouth pipettes for larval collection work.

Metal pillboxes, one-quarter ounce, for carrying adults captured.

Chloroform.

Formaldehyde.

Entomological forceps.

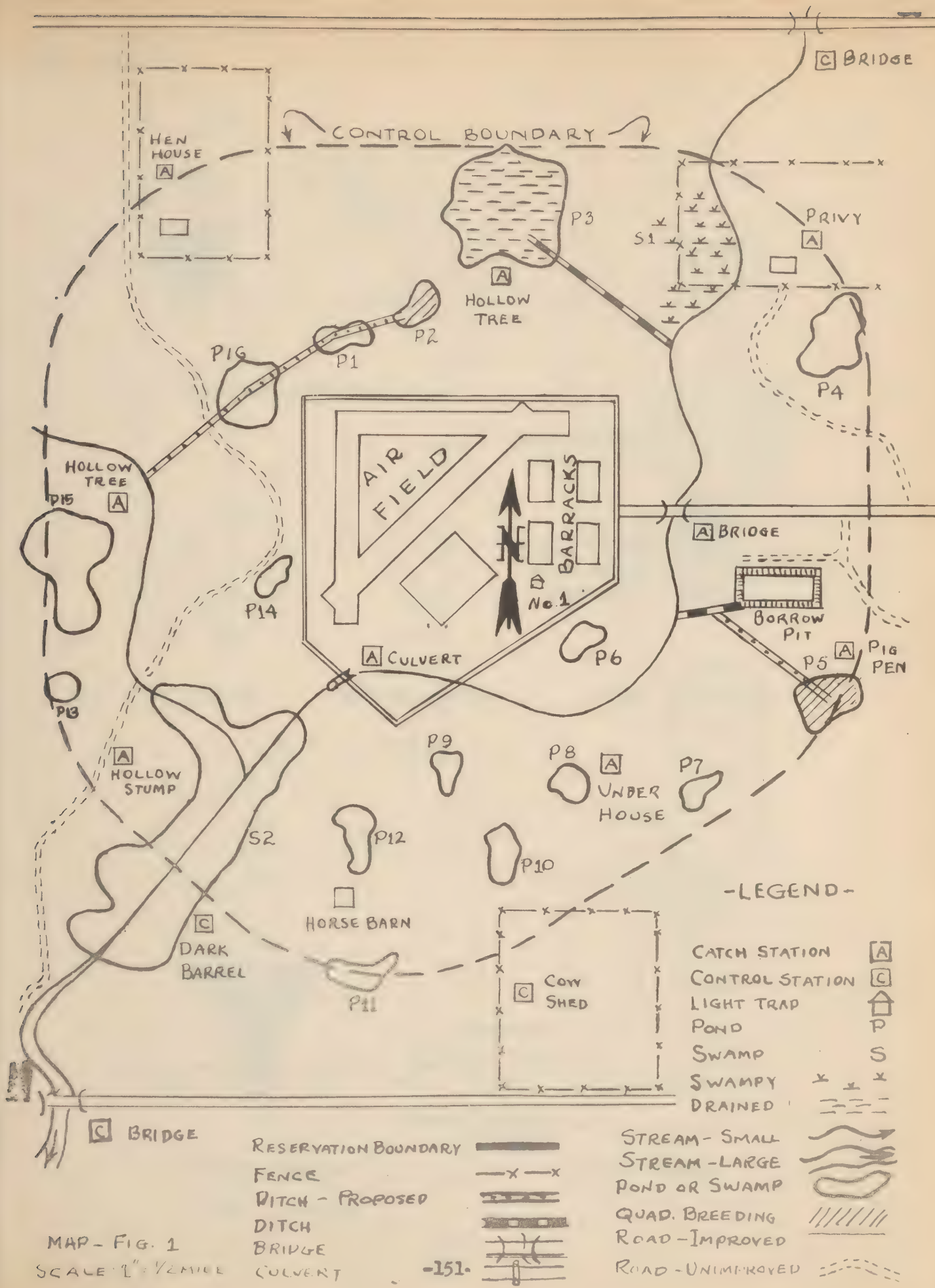
Dissecting needles.

Turtex belts for carrying glass larval vials.

Glass hanging drop slides.

Flashlights (three-cell) and extra batteries.

Dissecting binocular microscope (if procurable).



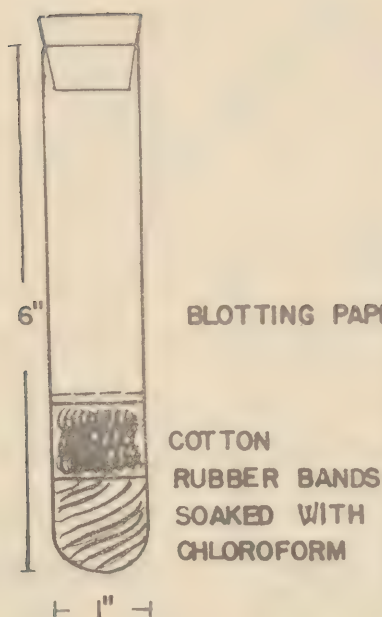


FIG. 2

CHLOROFORM TUBE

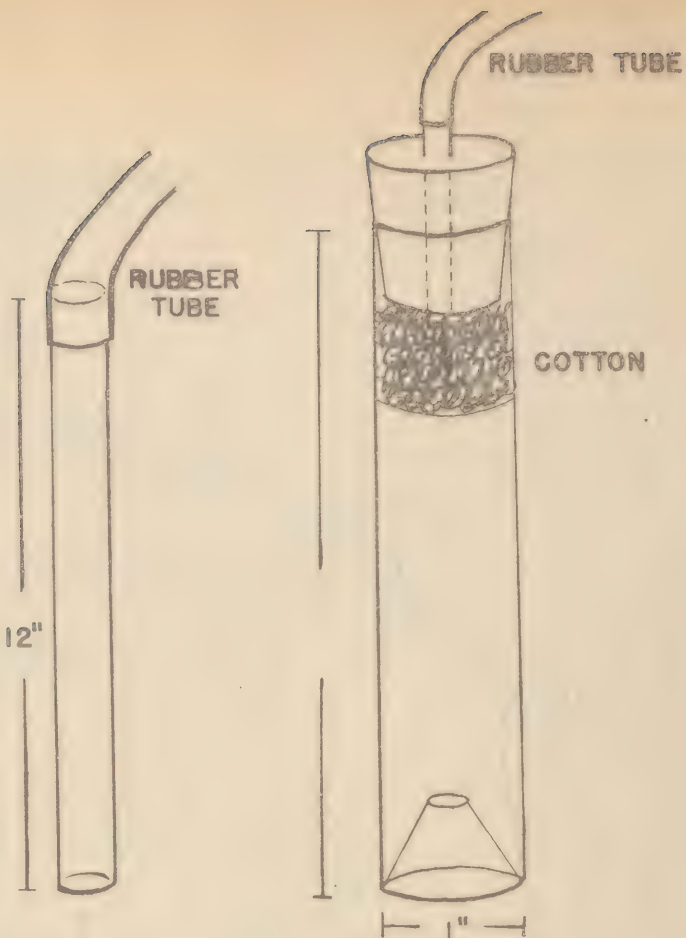
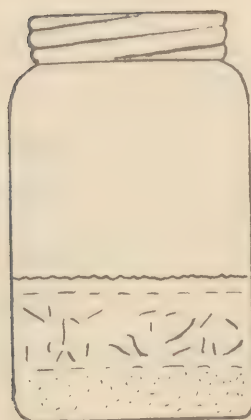


FIG. 3

LONG GLASS TUBE

SPECIAL TYPE TUBE

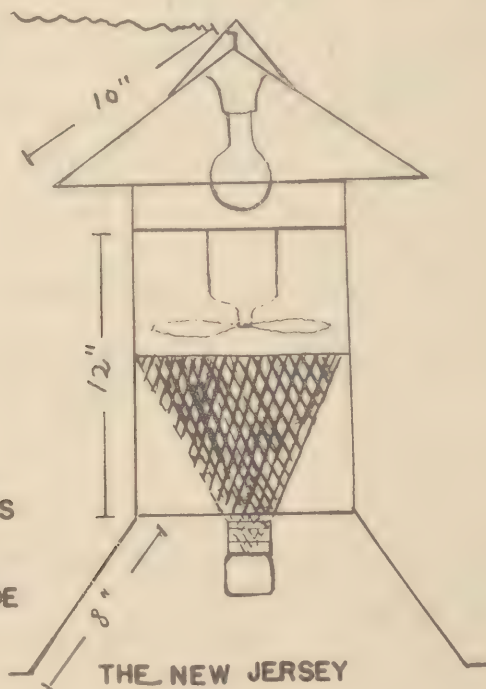


CYANIDE BOTTLE

CARDBOARD

WOOD SHAVINGS

SODIUM CYANIDE CRYSTALS



THE NEW JERSEY MOSQUITO TRAP

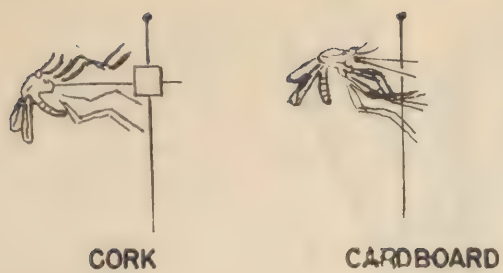


FIG. 7 MOUNTING MOSQUITOES

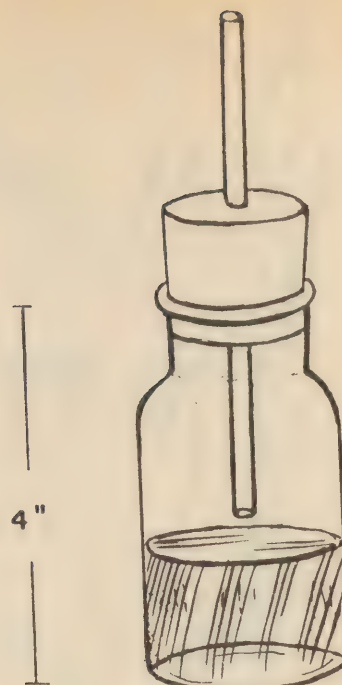


FIG. 8  
LARVAE SHIPPING BOTTLE

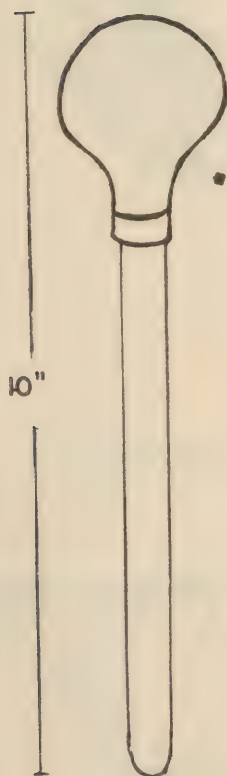


FIG. 9  
LARGE PIPETTE

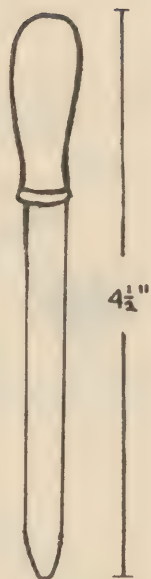


FIG. 10  
SMALL PIPETTE

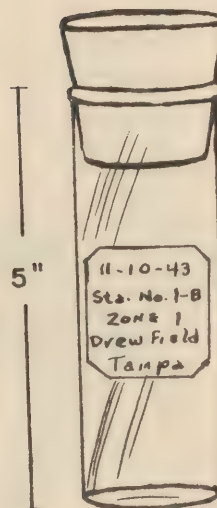


FIG. 11  
PLASTIC VIAL

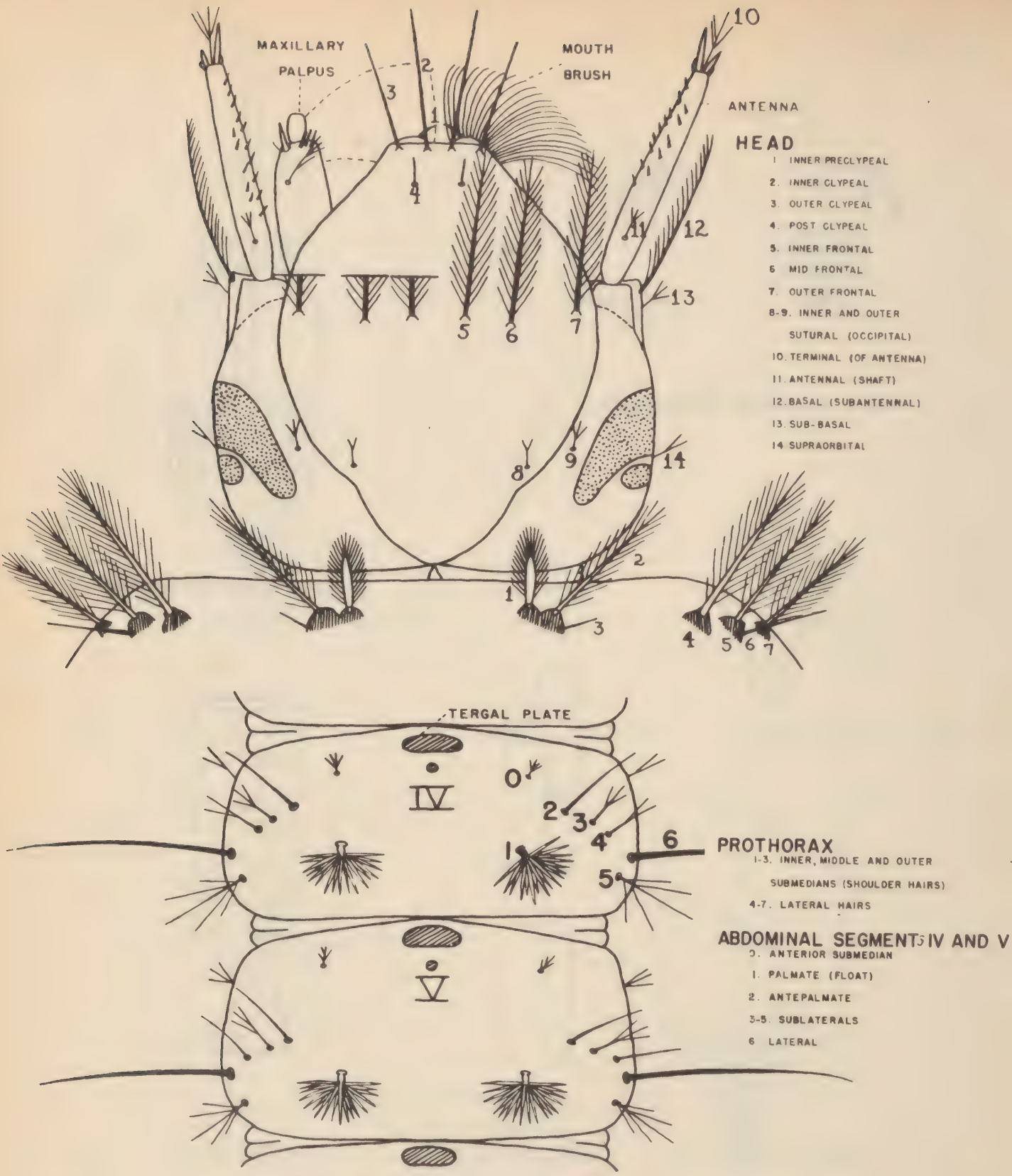
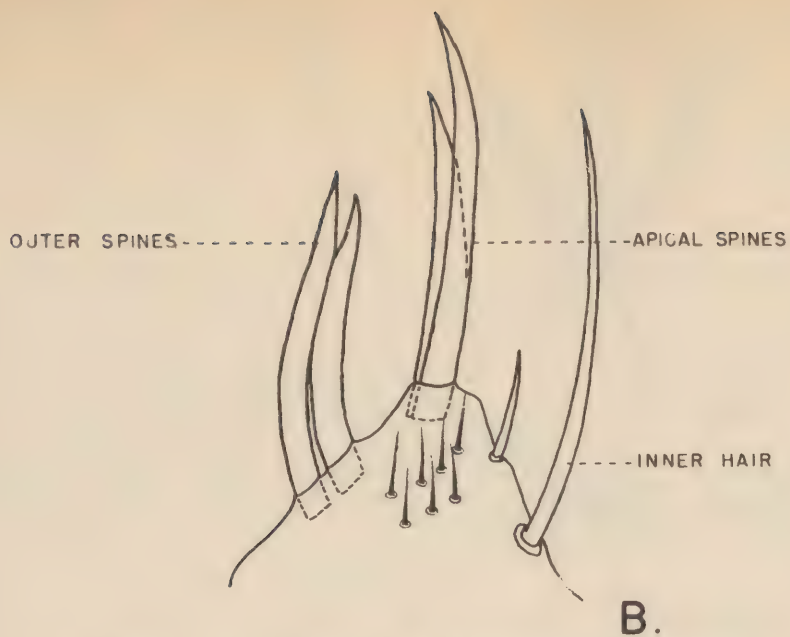
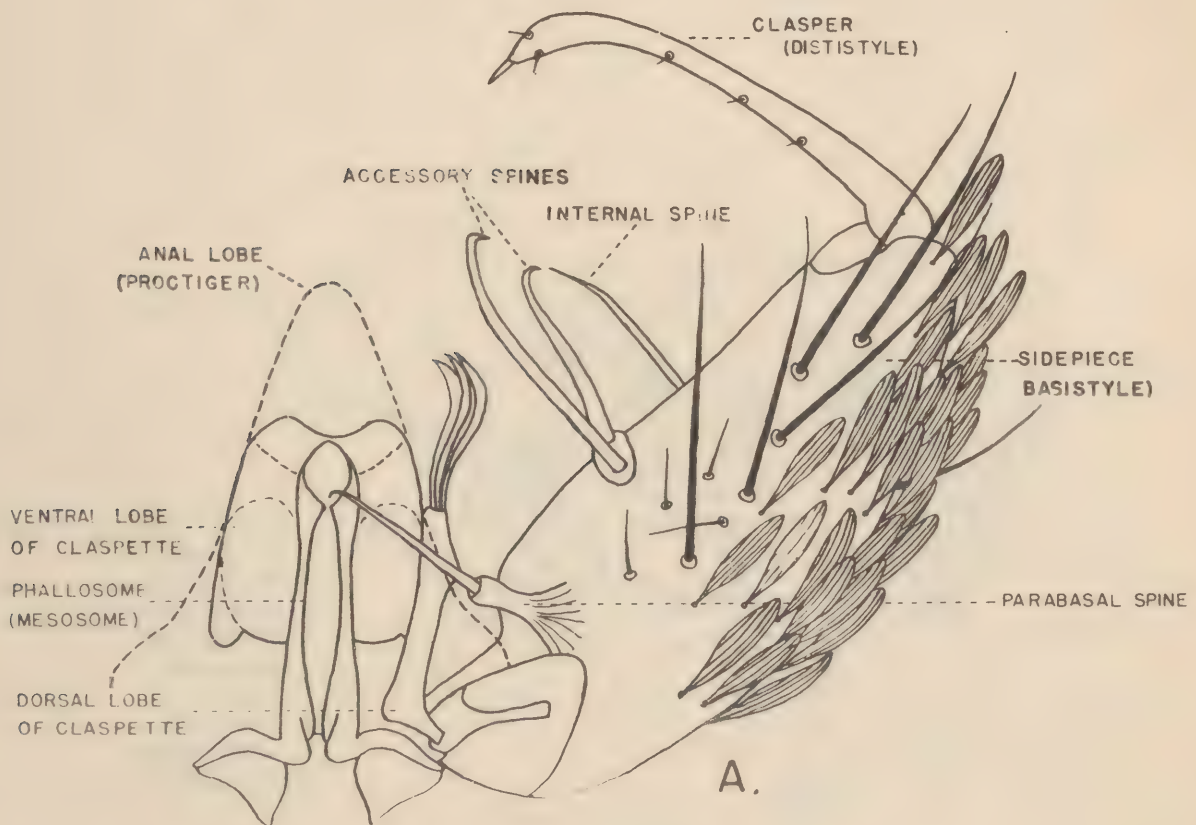


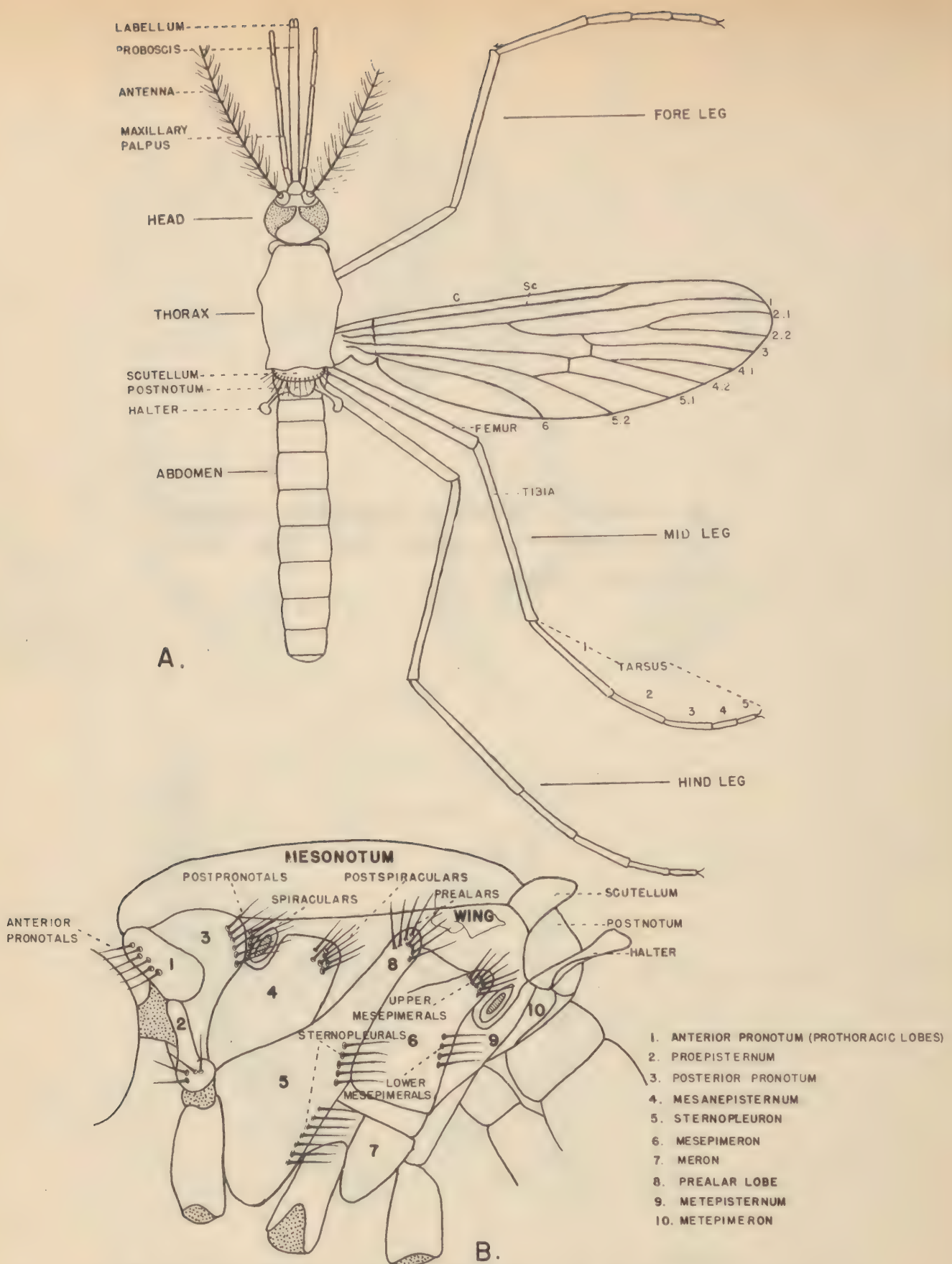
Diagram of the head and abdominal segments IV and V of Anopheles albimanus, showing conventional names and numbers of larval hairs.



B. Claspette spines of Anopheles crucians Wied. (After King. Amer. Jour. Trop. Med. 19: 461, 1939).



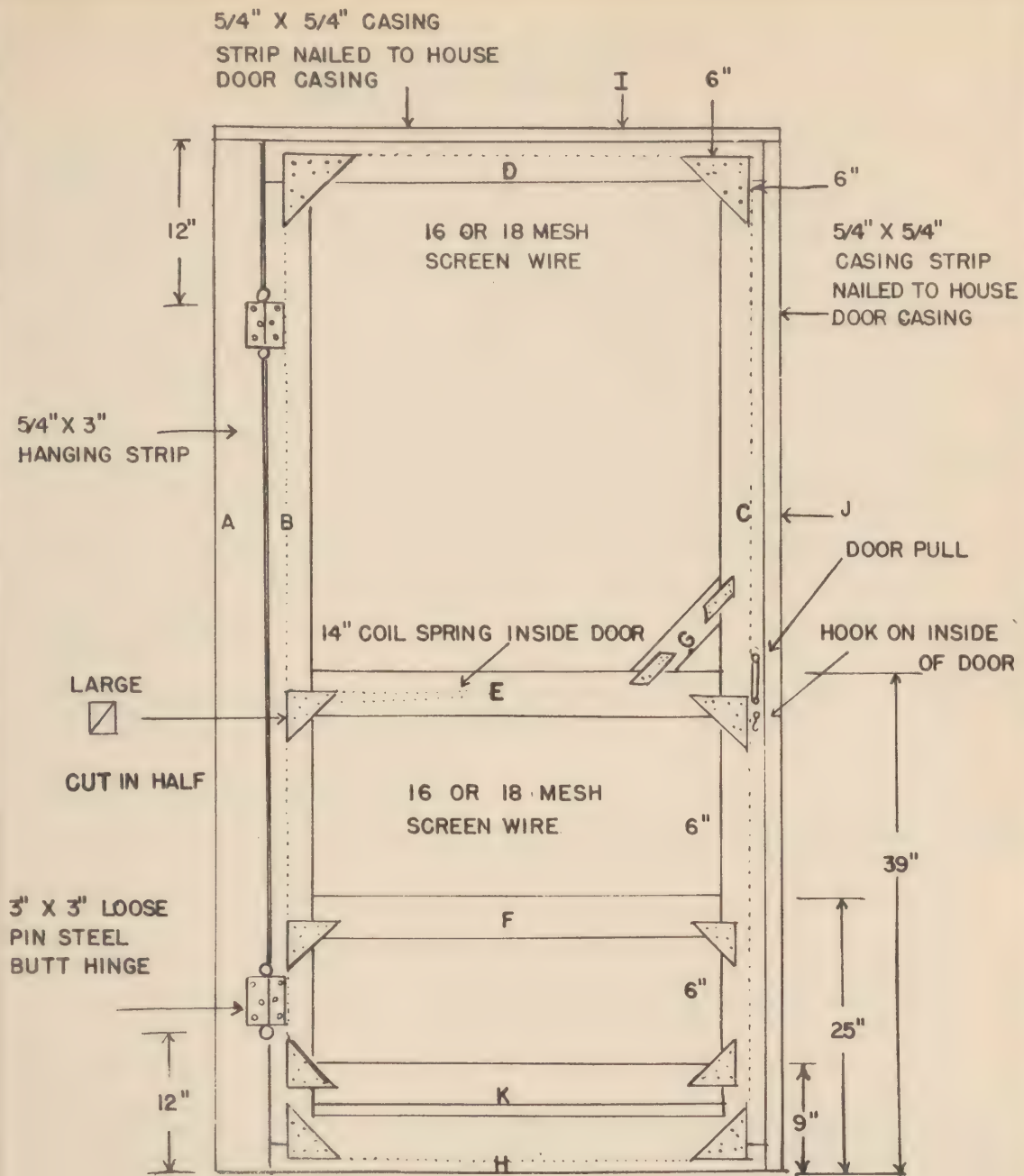
A. Male terminalia of Anopheles albimanus Wied. (Ninth tergite removed )



A. Generalized diagram of a (female) mosquito, to show nomenclature of parts. B. Lateral aspect of a mosquito thorax to show principal parts and arrangement of pleural setae.

MOSQUITO PROOFING





DOOR FRAME EXCEPT H IS 5/4" X 3" NO.1 CYPRESS OR PINE

H IS 5/4" X 6" NO.1 CYPRESS OR PINE

HANGING STRIP IS 5/4" X 3" NO.1 CYPRESS OR PINE

CASING STRIP IS 5/4" X 5/4" STOCK MATERIAL

ESCAMBIA COUNTY HEALTH DEPT.

J.E. ELMENDORF, M.D., DIRECTOR  
DAVID B. LEE, SANITARY ENGINEER

J. A. MULRENNAN, ENTOMOLOGIST



SPECIFICATION OF MATERIALS  
FOR MOSQUITO PROOFING

1. LUMBER

Shall be thoroughly dried clear cypress or pine; grade of B or better or No. 1.

- a. Screen door frames, 5/4" x 3", D-4-S, and 5/4" x 6", D-4-S.
- b. Screen door hanging strip, 5/4" x 3", D-4-S.
- c. Casing strip, stock 5/4" 5/4".
- d. Door facings and window facings, flooring, ceiling, etc.--

Shall be No. 1 pine, size specified on survey sheets.  
See bill of materials for average door.

2. SCREEN WIRES

Shall be galvanized wire screen preferably of 18 meshes per inch, though 16 meshes per inch can be used.

3. CONSTRUCTION OF SCREEN DOORS

The attached drawing shall be followed in constructing screen doors.

4. MATERIALS FOR SCREEN DOORS

- a. Reinforcing Plates for Screen Doors.

To be cut from squares of 28 gauge galvanized sheet metal.

- b. Hardware cloth.

The screen wire in the bottom panel of screen doors may be reinforced with galvanized hardware cloth, 4 x 4 meshes per inch standard gauge wire. This is advisable, but expense, at times, prohibits its use.

- c. Nails.

The wire clout nails used in fastening metal reinforcing plates on the outside shall be 1" length (2nd common). The inside reinforcing plates shall be put on over the screen wire and fastened with wire clout nails of length equal to the thickness of the wood frame.

d. Hinges.

The hinges shall be 3" x 3" steel loose pin, butt-type and fastened with #8, 1" screws.

e. Other Items.

One door pull, hook and eye, and one 14" coil spring, corrugated fasteners.

5. APPLICATION OF WINDOW SCREENS

Window screens, when wire is tacked directly on frames, shall overlap the window frames 1" on top and sides. The bottom edge shall be overlapped 2" on the sill, and the wire securely fastened with a double row of #6 bill poster tacks, spaced 1" apart.

6. MOSQUITO PROOFING MATERIAL

a. Paper.

The paper for covering walls and ceiling shall be 90# basis kraft weighing not less than 1 pound per 34 square feet.

b. Tacks.

The paper shall be securely fastened to the walls with #6 bill poster tacks.

c. Roofing Paper.

The paper specified for covering floors shall be two-ply roofing paper weighing about 35 pounds per square.

d. Sheet Metal.

The sheet metal for patching, as shown on the survey sheet, shall be twenty-eight gauge galvanized sheet metal.

# HARDWARE

Name:  
Address:  
Deliver to:

- \_\_\_ pr. 3" x 3" steel, loose pin, butt hinges and No. 8, 1" screws
- \_\_\_ 14" Coil Spring
- \_\_\_ Door pull
- \_\_\_ Hook and eye
- \_\_\_ Square foot of No. 28 gauge galvanized metal
- \_\_\_ lbs. 1" lathe nails or 2d common
- \_\_\_ lbs No. 6 bill poster tacks
- \_\_\_ lbs. No. 6 finishing nails
- \_\_\_ pts. paint (grey) (black)
- \_\_\_ lbs. No. 6d common nails
- \_\_\_ lbs. No. 4d common nails
- \_\_\_ Screen hanging sets
- \_\_\_ Lin. ft. of 16 or 18 mesh first grade galv. iron screen wire 24" wide
- \_\_\_ " " " " " " " " " " " " " " 26" "
- \_\_\_ " " " " " " " " " " " " " " 28" "
- \_\_\_ " " " " " " " " " " " " " " 30" "
- \_\_\_ " " " " " " " " " " " " " " 32" "
- \_\_\_ " " " " " " " " " " " " " " 34" "
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- \_\_\_ " " " " " " " " " " " " " " 40" "
- \_\_\_ " " " " " " " " " " " " " " 42" "

## LUMBER

Name:  
Address:  
Deliver to:

All specifications must be complied with to the letter.

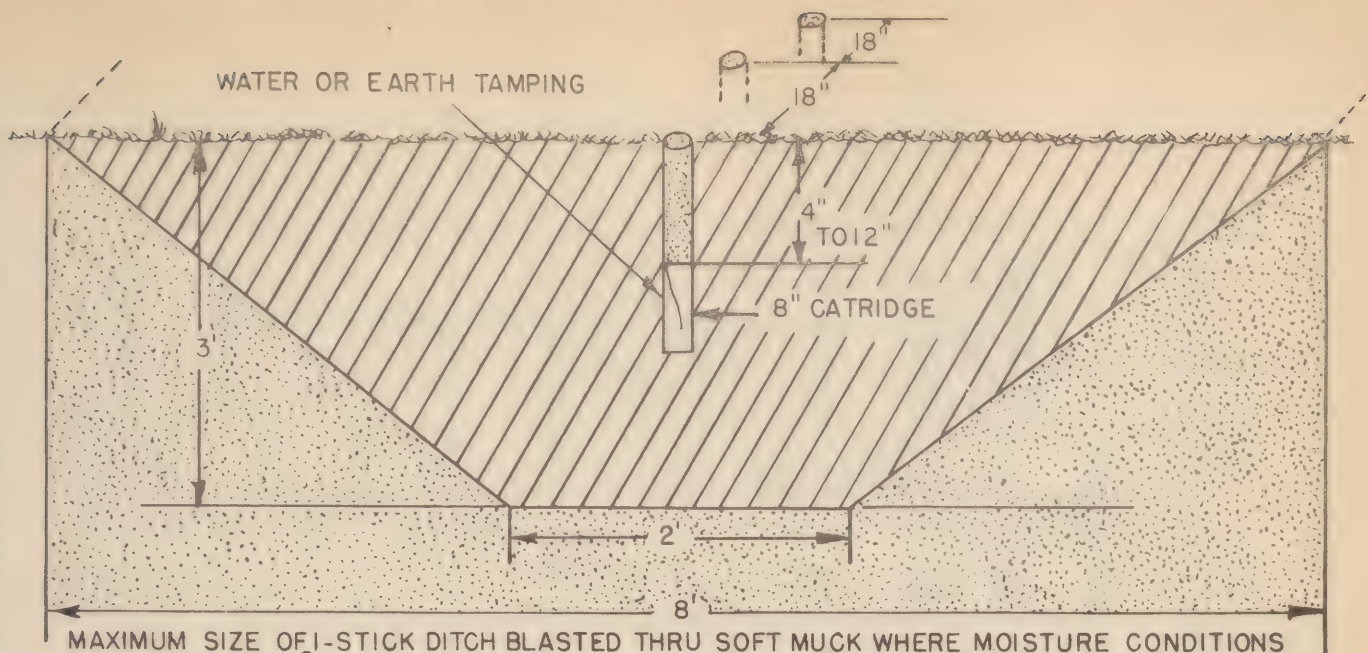
Lumber must be straight and 1st grade.

\_\_\_ pcs. 5/4" x 3" x 14' #1 cypress or pine D-4-S A.E.F.  
\_\_\_ pcs. 5/4" x 3" x 12' #1 cypress or pine D-4-S B.D.G.  
\_\_\_ pcs. 5/4" x 3" x 10' #1 cypress or pine D-4-S C.K.  
\_\_\_ pcs. 5/4" x 6" x 3½' #1 cypress or pine D-4-S H.  
\_\_\_ pcs. 5/4" x 5/4" x 10' #1 pine (stock) D-4-S I.J.  
\_\_\_ pcs. ½" x 1½" x #1 pine D-4-S  
\_\_\_ pcs. 1" x 2" x #1 pine D-4-S  
\_\_\_ pcs. 1" x 2" x #1 pine D-4-S  
\_\_\_ pcs. 1" x 3" x #1 pine D-4-S  
\_\_\_ pcs. 1" x 3" x #1 pine D-4-S  
\_\_\_ pcs. 1" x 4" x #1 pine D-4-S  
\_\_\_ pcs. 1" x 4" x #1 pine D-4-S  
\_\_\_ pcs. 1" x 6" x #1 pine D-4-S  
\_\_\_ pcs. 1" x 6" x #1 pine D-4-S  
\_\_\_ pcs. 2" x 4" x #1 pine D-4-S  
\_\_\_ pcs. 2" x 4" x #1 pine D-4-S  
\_\_\_ rolls single ply roofing

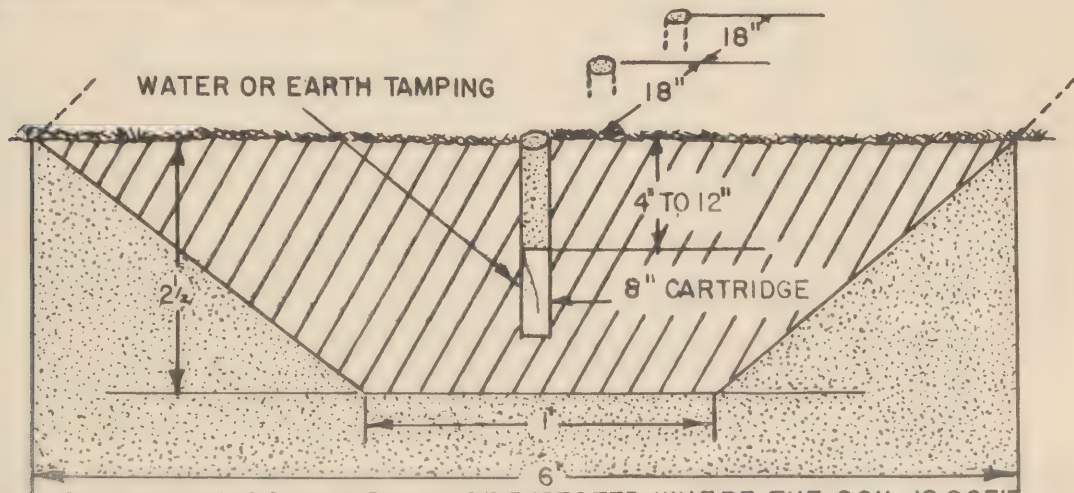
These specifications should be complied with wherever possible.

The first five items listed cover pieces of lumber for manufacture of one door. The remaining items are listed for work of repair to house before mosquito-proofing.

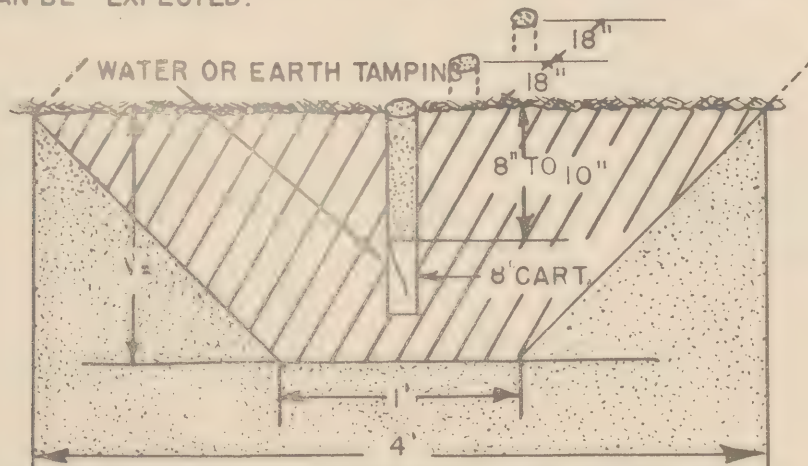
# PROPAGATION DITCHES—EXPECTED SHAPE AND SIZE



MAXIMUM SIZE OF I-STICK DITCH BLASTED THRU SOFT MUCK WHERE MOISTURE CONDITIONS ARE IDEAL UP TO  $1\frac{2}{3}$  CU.YDS. OF MATERIAL PER POUND OF 50% NITROGLYCERIN DYNAMITE MAY RESULT UNDER SUCH CONDITIONS.



USUAL SIZE I-STICK DITCH TO BE EXPECTED WHERE THE SOIL IS SOFT AND WET. REMOVAL OF ONE CU.YD. OF MATERIAL PER POUND OF 50% NITROGLYCERIN DYNAMITE CAN BE EXPECTED.



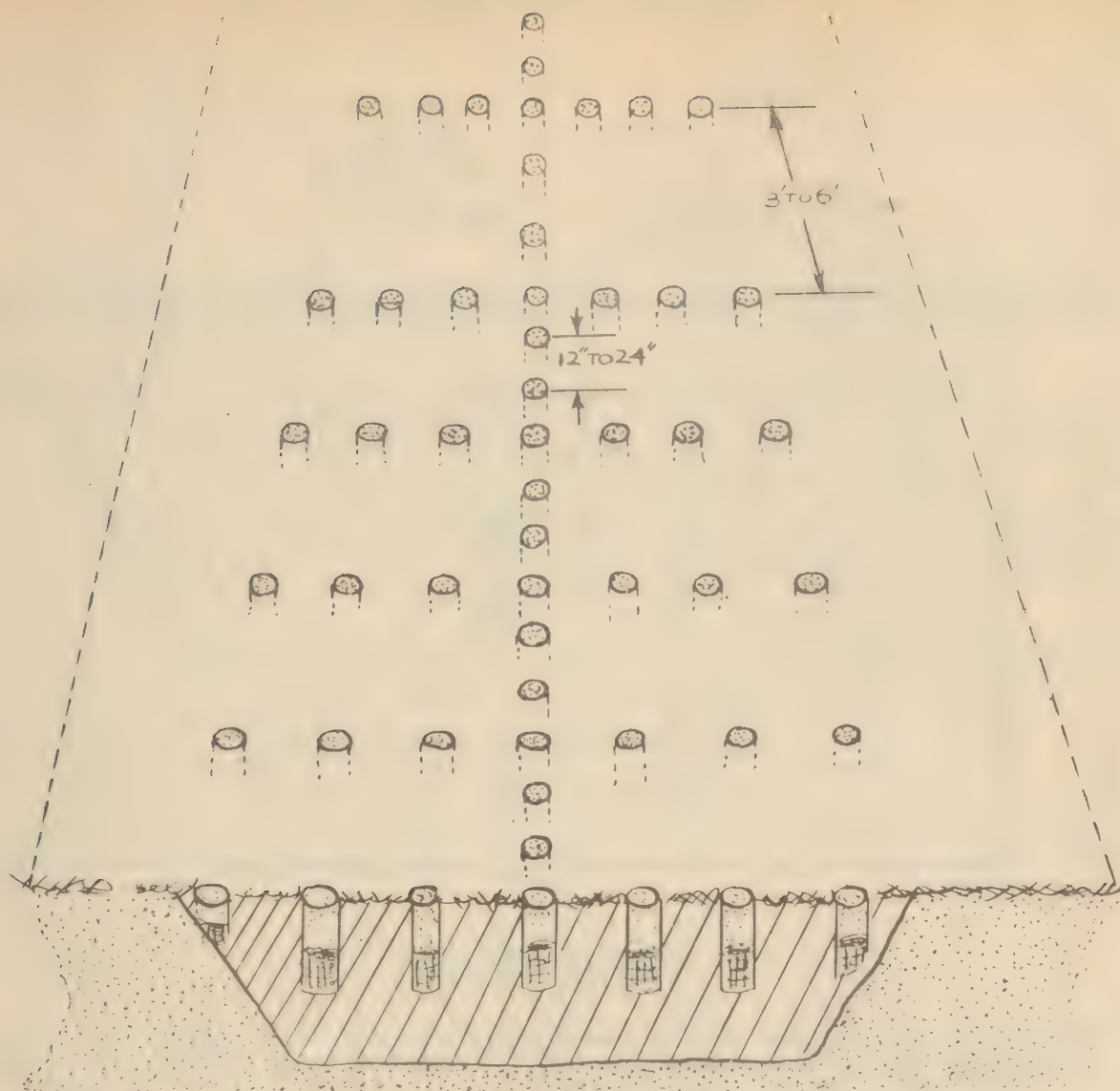
MINIMUM SIZE OF I-STICK DITCH THRU HEAVY SOIL AND WITH MINIMUM MOISTURE CONDITIONS FOR PROPAGATION. ONE HALF CU.YD. OF MATERIAL REMOVED UNDER THESE CIRCUMSTANCES IS TO BE EXPECTED.

# CHARGES OF 50% STRAIGHT DYNAMITE

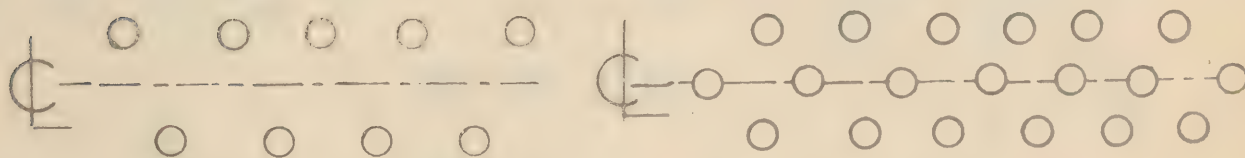
## FOR BLASTING DITCHES BY PROPAGATION METHOD

TOP WIDTH OF DITCH ft.	APPROXIMATE NO. OF 1 $\frac{1}{4}$ " X 8" CARTRIDGES PER HOLE REQUIRED FOR VARIOUS DEPTHS				NUMBER OF PARALLEL ROWS REQUIRED	DISTANCE BETWEEN ROWS IN INCHES
	2 $\frac{1}{2}$ to 3 ft.	4 ft.	5 ft.	6 ft.		
6	1	2	5	6	1	"
8	1	2	3	"	1 or 2	30
10	1	2	3	5	2	36
12	1	2	3	5	2	42
14	1	2	3	5	2	48
16	1	2	3	5	3	36
18	1	2	3	5	3	42

THE CHARGES ABOVE ARE APPROXIMATE FOR HOLES SPACED 24 INCHES APART—THE EXACT SPACING DETERMINED BY 3 OR 4 TEST SHOTS.



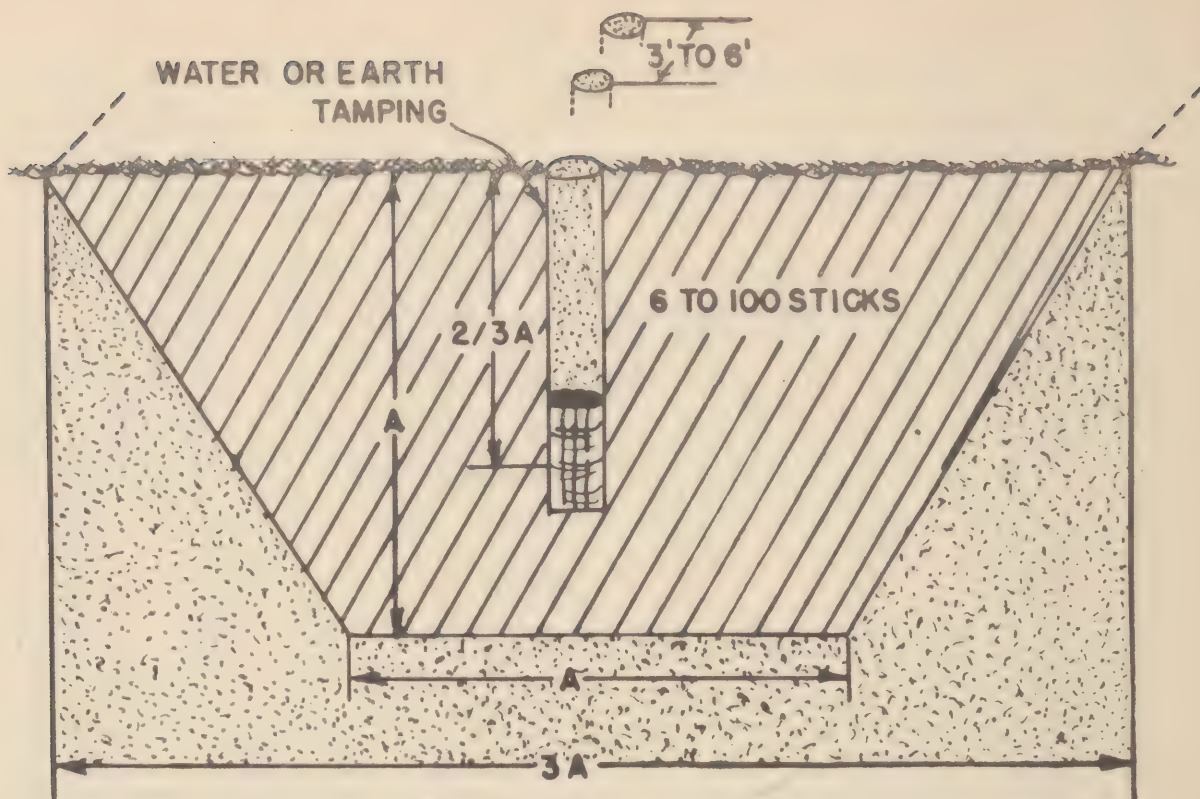
CROSS SECTION METHOD OF DYNAMITING DITCHES



— PARALLEL LINE METHOD —

AFTER DU PONT'S BLASTERS HAND BOOK ELEVENTH EDITION

# DITCHING POST HOLE METHOD

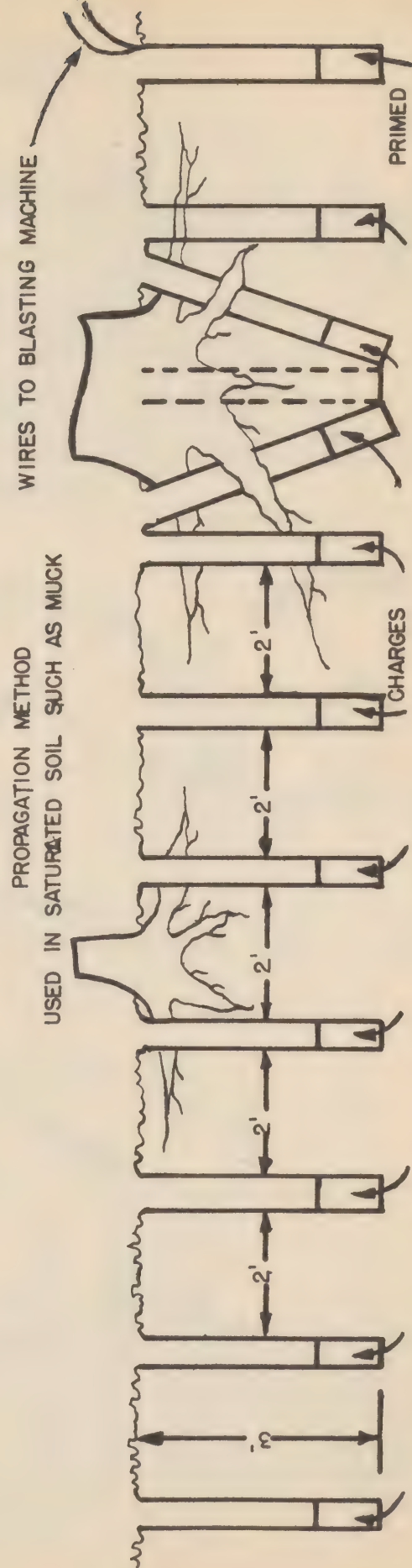
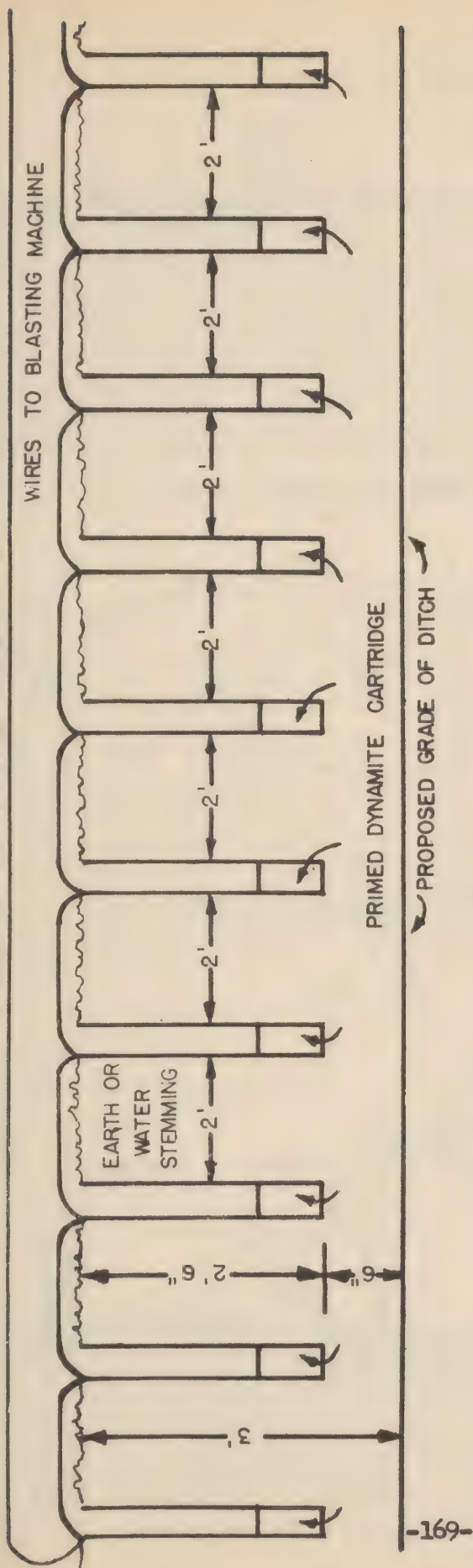


## SPECIFICATIONS

NUMBER OF STICKS ( $1\frac{1}{4} \times 8''$ ) PER HOLE .....	6	10	20	30	50	100
NUMBER OF POUNDS PER HOLE .....	3	5	10	15	25	50
DISTANCE BETWEEN HOLES (FEET) .....	3	$3\frac{1}{2}$	4	$4\frac{1}{2}$	5	6
DEPTH OF DITCH ("A" FEET) .....	4	5	6	7	$8\frac{1}{2}$	12
BOTTOM WIDTH OF DITCH ("A" FEET) .....	4	5	6	7	$8\frac{1}{2}$	12
TOP WIDTH OF DITCH ("3A" FEET) .....	12	15	18	21	$25\frac{1}{2}$	36
DEPTH OF LOAD ( $\frac{2}{3}$ OF "A" FEET) .....	$2\frac{2}{3}$	$3\frac{1}{3}$	4	$4\frac{2}{3}$	$5\frac{1}{2}$	8
DIAMETER OF POST HOLE (INCHES) .....	4	4	6	6	8	8
DYNAMITE PER. 100' OF DITCH .....	100	$142\frac{1}{2}$	230	333	500	833
MATERIALS MOVED PER. 100' (CU. YDS.) .....	118	185	266	363	533	1067

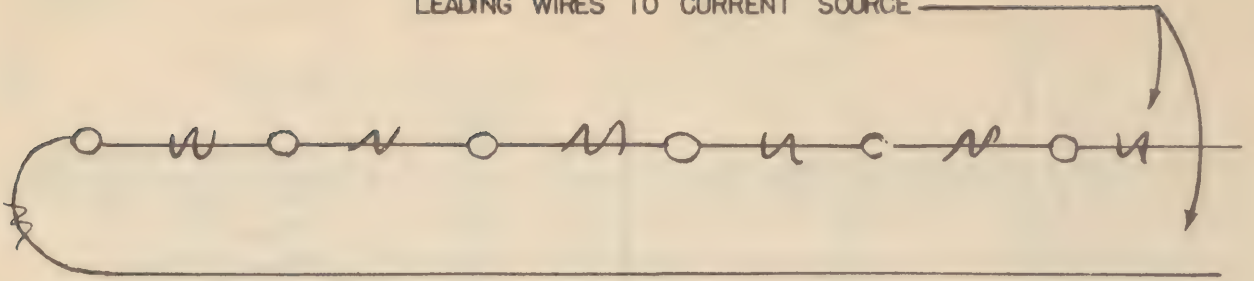
## AFTER DU PONT

# DITCHING ELECTRIC METHOD USED IN DRY SAND OR HEAVY CLAY SOIL

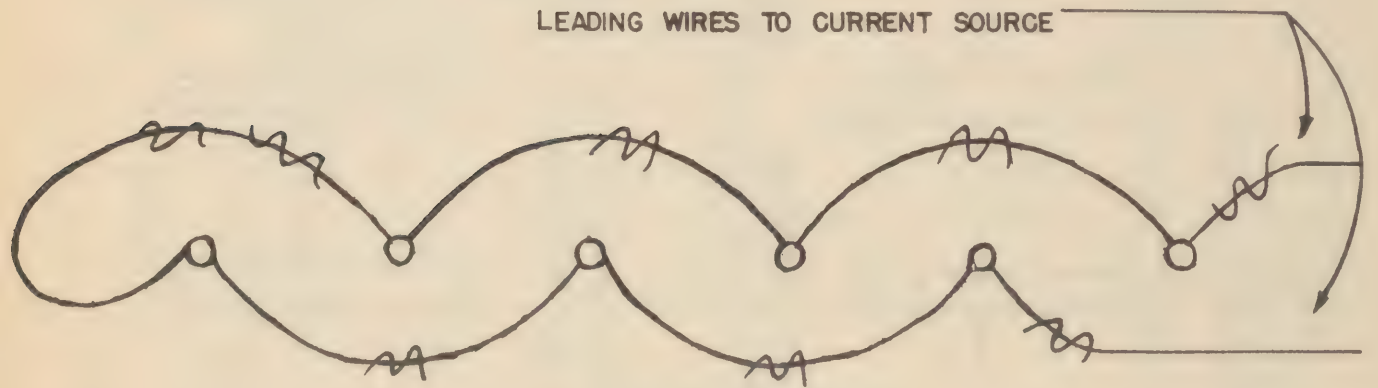


# FIRING ELECTRICALLY

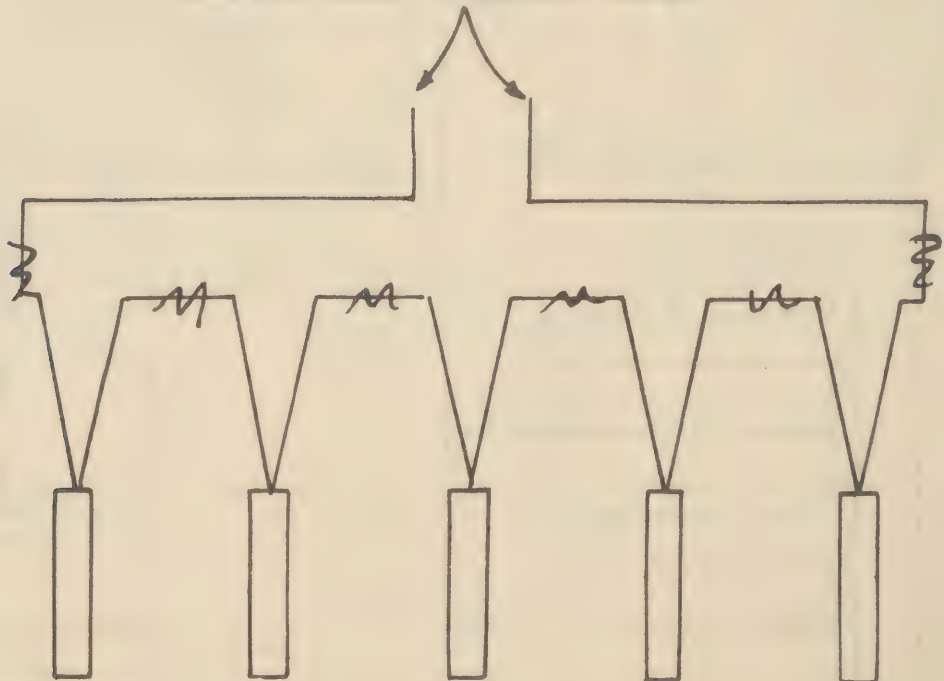
LEADING WIRES TO CURRENT SOURCE



LEADING WIRES TO CURRENT SOURCE



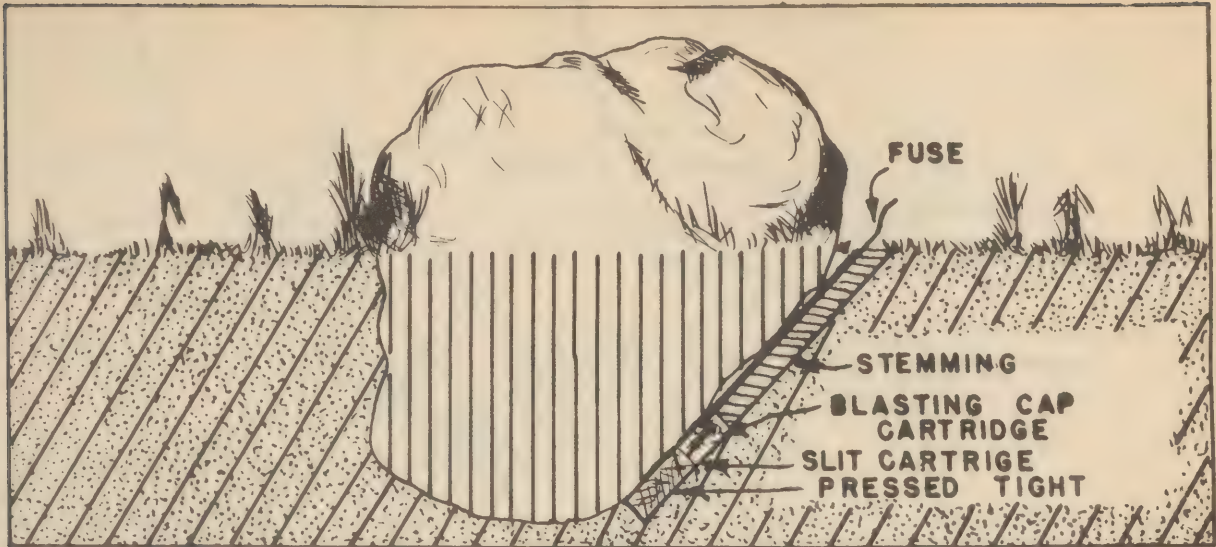
LEADING WIRES TO CURRENT SOURCE



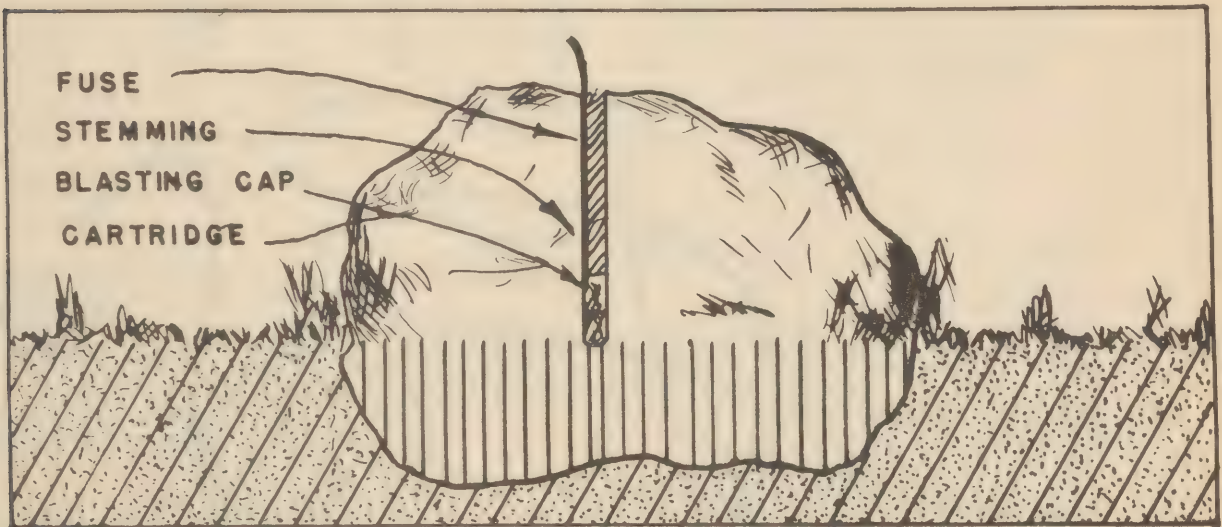
AFTER DUPONT  
BLASTERS' HANDBOOK, ELEVENTH EDITION.

# BOULDER — BLASTING

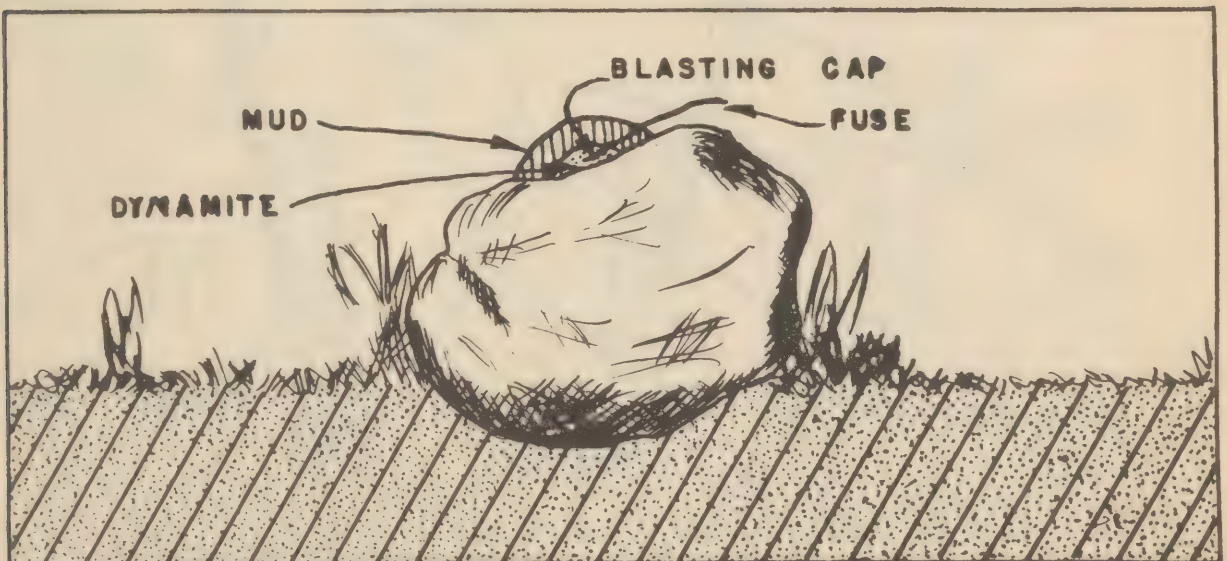
— SNAKE HOLE METHOD



BLOCK HOLE METHOD

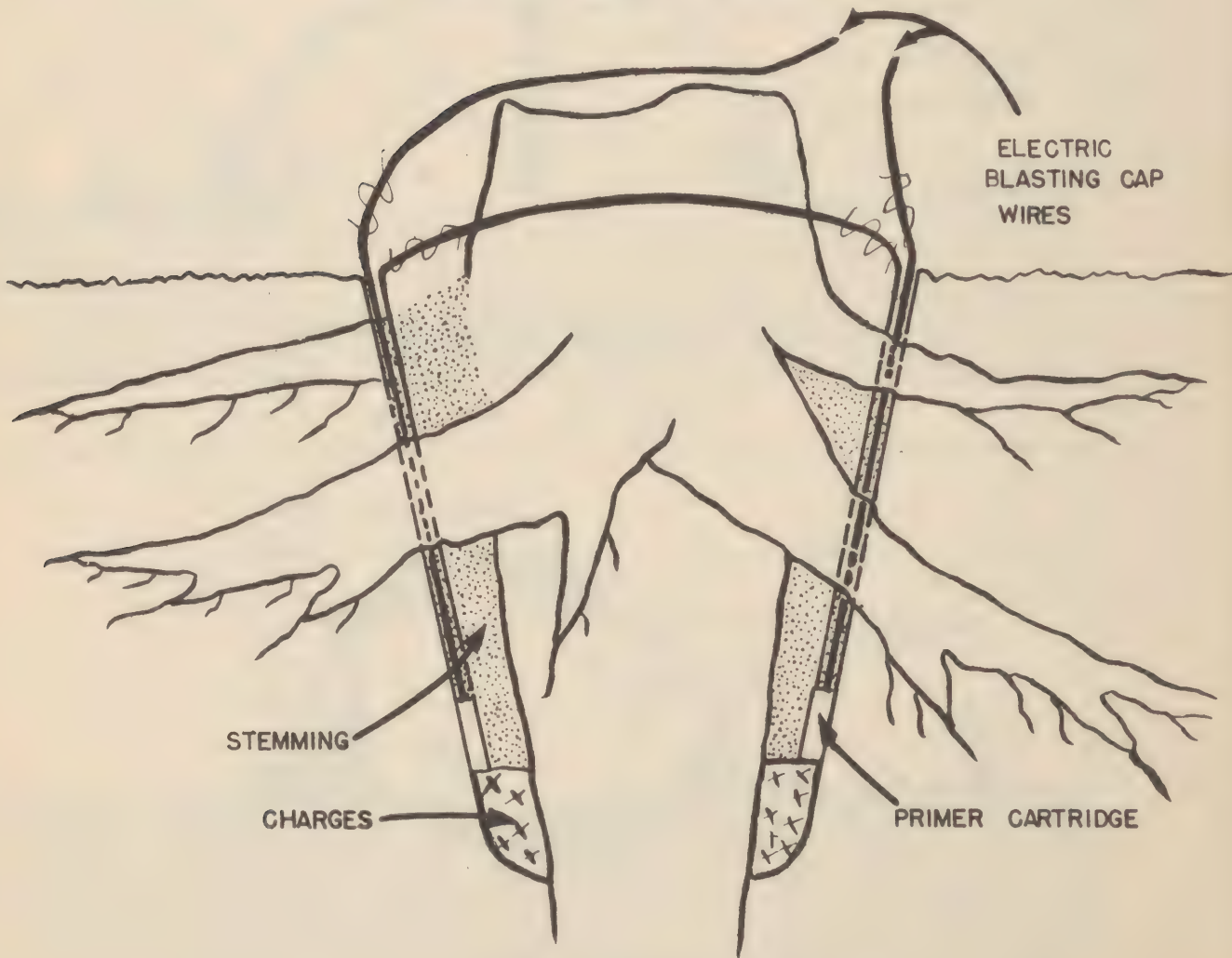
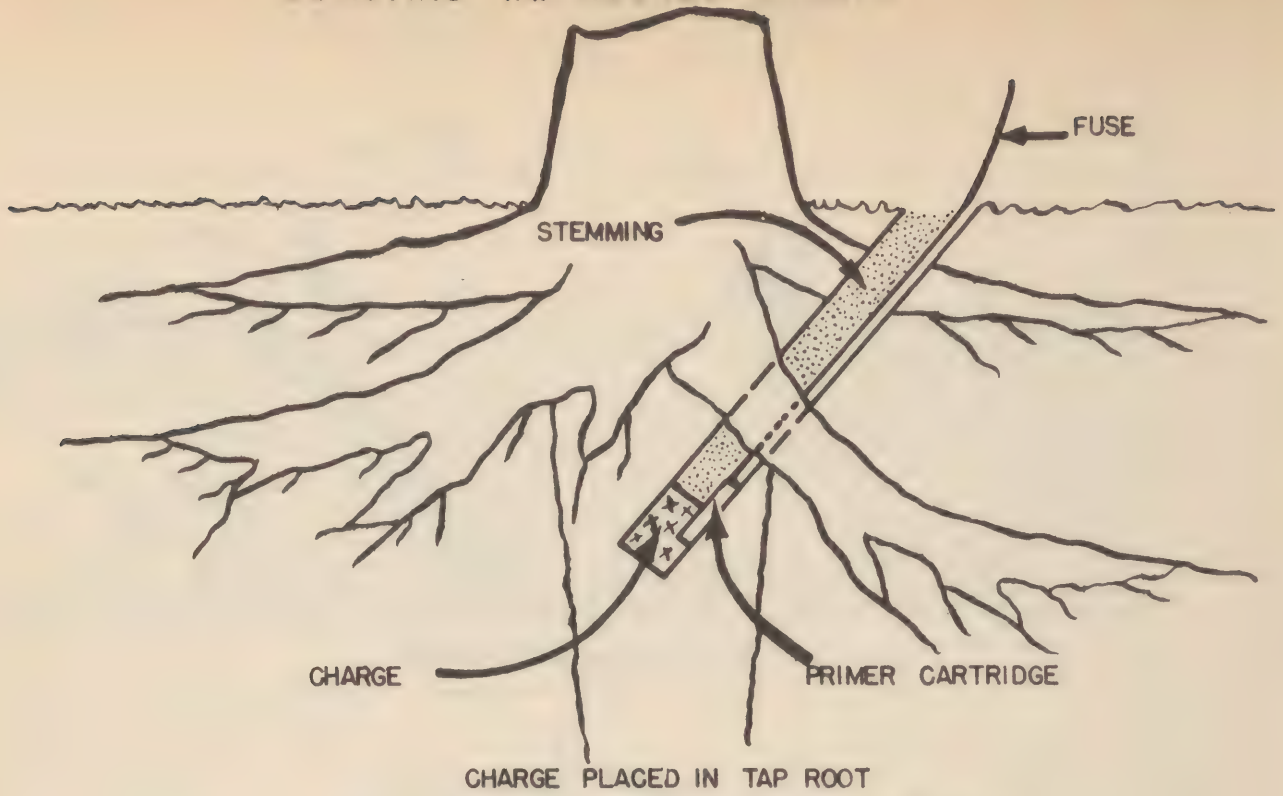


MUDCAP METHOD



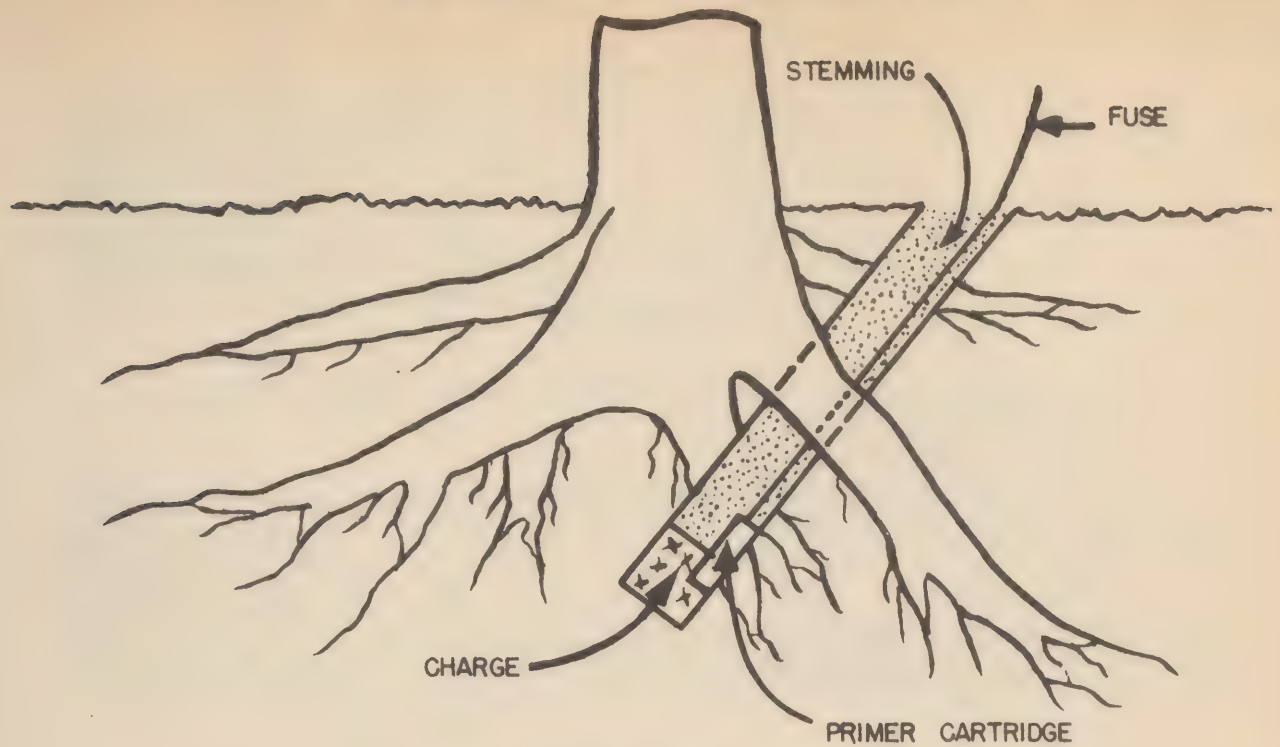
AFTER DUPONTS BLASTERS HAND BOOK ELEVENTH EDITION

# BLASTING TAP ROOTED STUMPS

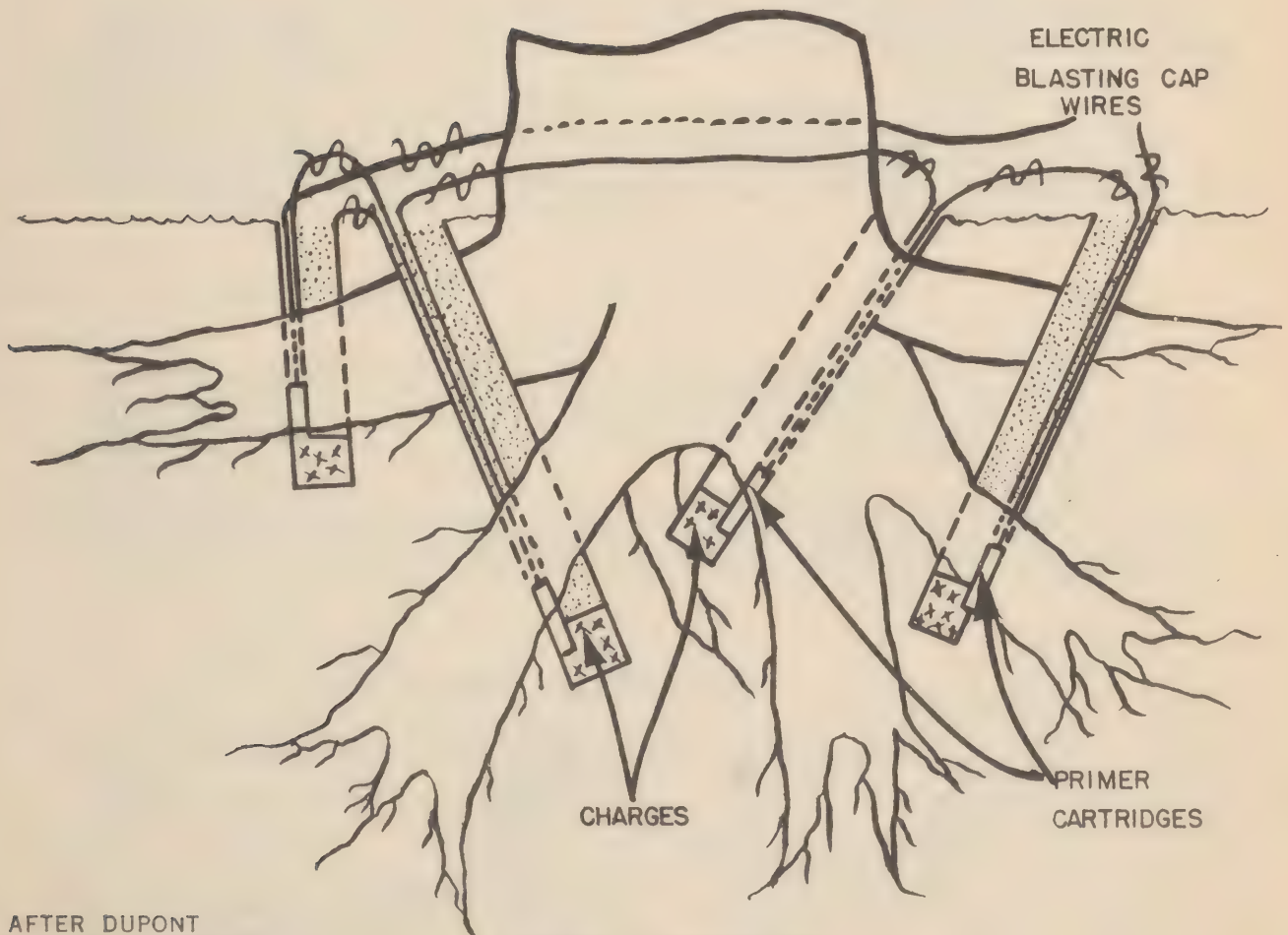


LOADING HOLES ALONGSIDE ROOT

# BLASTING LATERAL ROOTED STUMPS

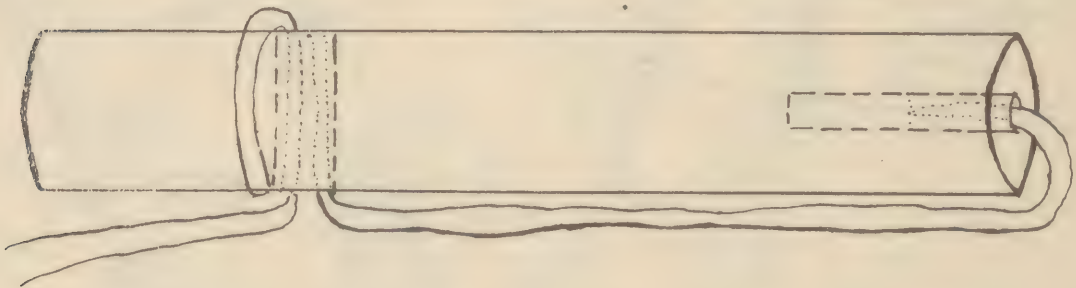
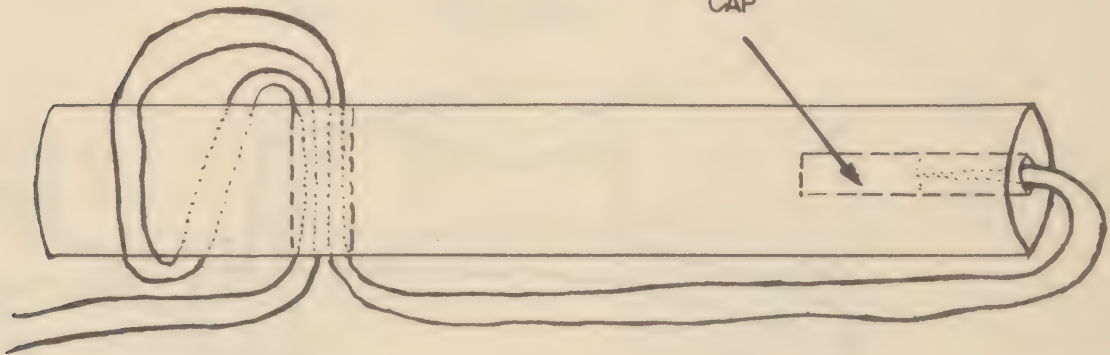
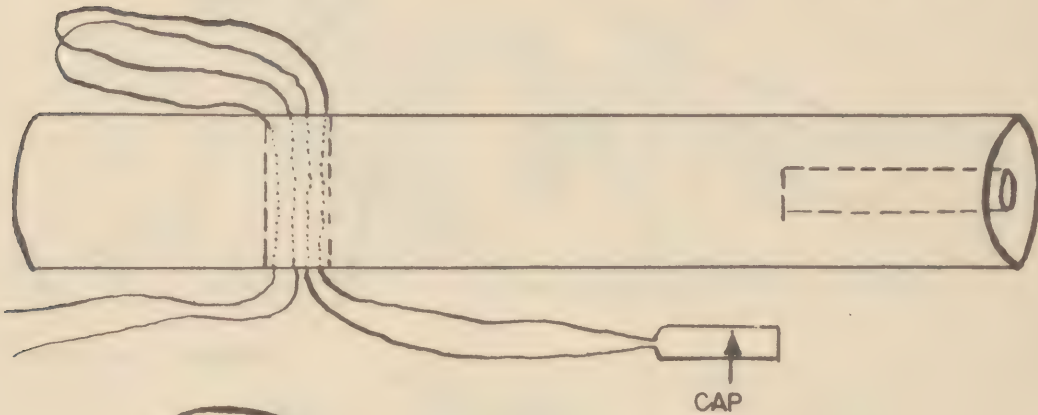
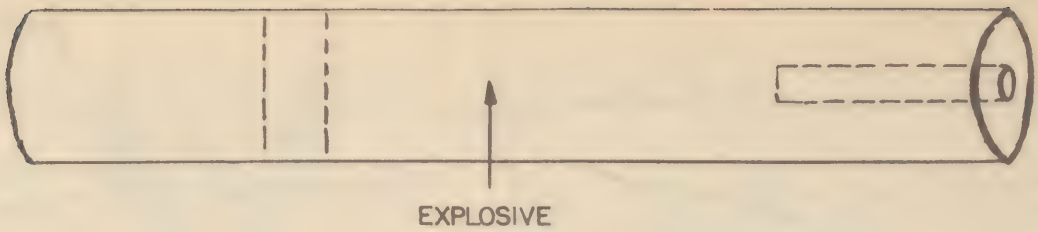


LOADING SMALL STUMP

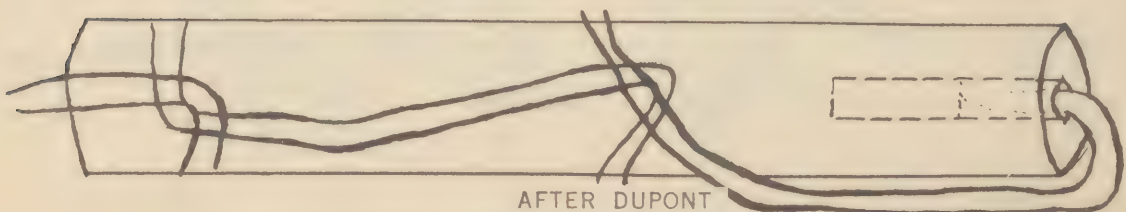


LOADING LARGE STUMP

ACCEPTED METHOD

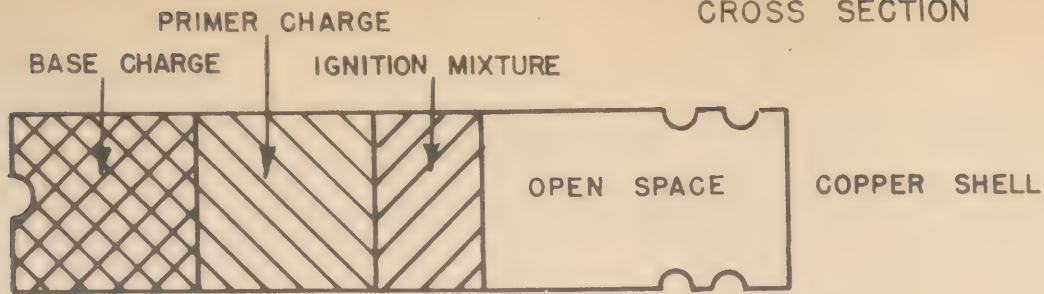


MOST COMMONLY USED METHOD

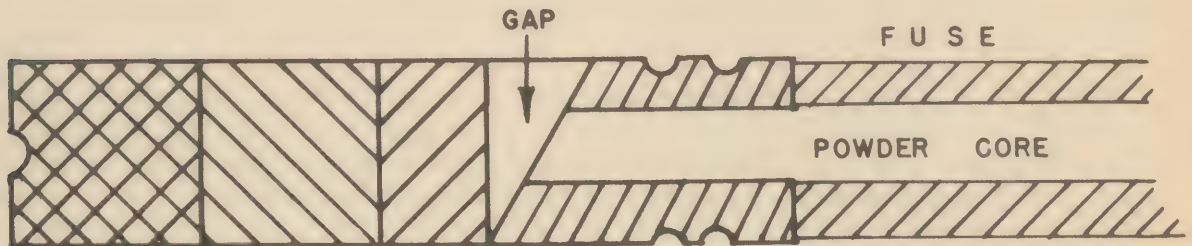


# BLASTING CAPS WITH SAFETY FUSE

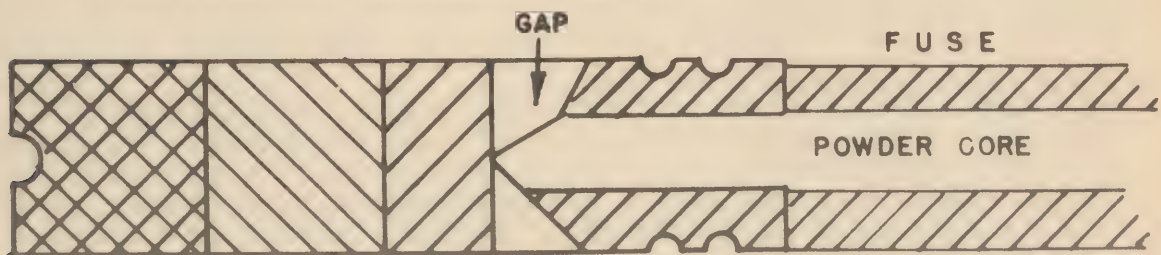
## CROSS SECTION



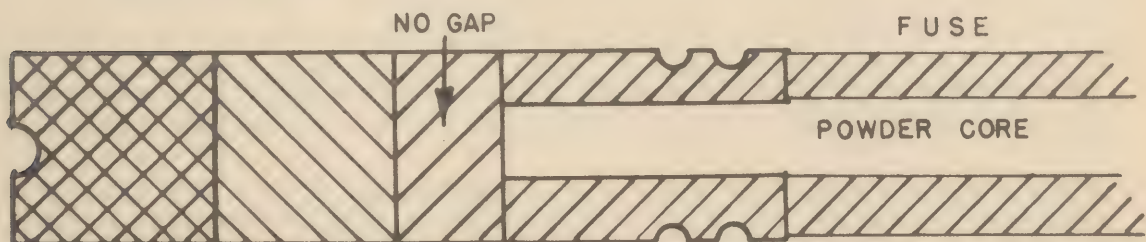
## SAFETY FUSE IMPROPERLY CUT



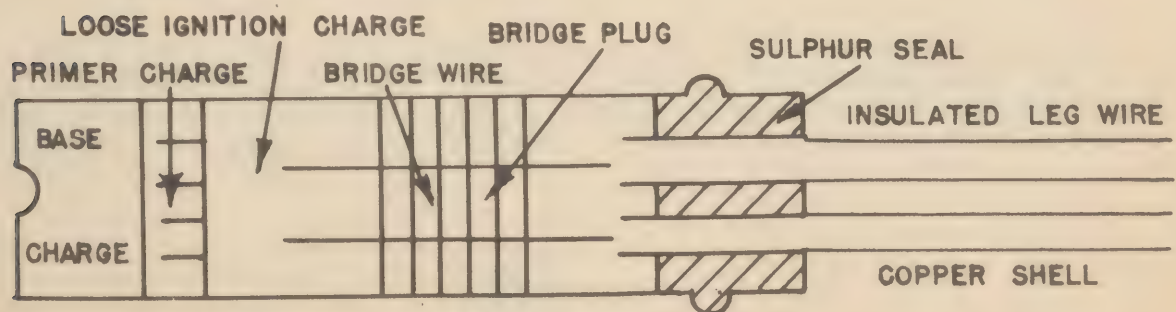
## SAFETY FUSE IMPROPERLY CUT



## SAFETY FUSE PROPERLY CUT



## ELECTRIC BLASTING CAP



AFTER DU PONT



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SKETCH MAPPING FOR PUBLIC HEALTH WORK

By

Fernald E. Hulse

(NOTE: This article was copied from that originally prepared by Fernald E. Hulse. If errors should exist in this reproduction, they are not attributable to the original author).

The public health officer of today recognizes the necessity of having good maps of the territory under his consideration. Especially is this true in yellow fever, malaria, and hookworm studies. It has been found, however, that it is not always possible to obtain reliable maps of all territories, especially in most foreign countries. Therefore, it is quite necessary that the public health officer, whether a physician or otherwise, should know the principles of sketch mapping in order to construct maps sufficiently accurate in scale and represented data to be of use in his work. It is the purpose of this paper to present the subject of sketch mapping in the simplest form possible, following closely the methods employed in the military service of the United States.

In preparing sketch maps of this nature no measuring instruments are necessary other than one's pace or stride. The pace or stride of the individual must, therefore, be accurately determined as the basic unit of measure. A pace is the distance from the heel of one foot to the heel of the other foot in walking naturally. A stride is the distance from the heel of one foot to the heel of the same foot as it strikes the ground the second time, or, in other words, a stride is equal to two paces. In order to measure one's pace or stride it will be necessary to lay off on the ground an accurately measured distance of say 500 yards; double this distance would be better. This distance should then be paced off at least four times, noting the number of paces each time. The average of these trials will give a fairly accurate result. For example, if the average were 510 paces for a distance of 500 yards, this would make 35.3 inches the actual length of one pace. A stride would be twice this or 70.6 inches.

Without going into the mathematics of converting this pace or stride into the scale desired on the map as inches to miles or inches to feet, charts P and S may be consulted and the corresponding scales to the figured length of pace or stride is that to be used in plotting the map in the field.

The ordinary scales adopted for use in the military service are 3 inches, 6 inches, or 12 inches to 1 mile. However, by using these scales in uneven feet, it will be found more difficult to reproduce enlargements of the field sketch when the case arises. In the engineering profession the

ordinary scale for purposes of enlargement can be easily handled. Hence the scales here chosen for the field or flat sketch maps are 1 inch to 1,600 feet and 1 inch to 800 feet. On the basis of these two scales Charts P and S have been plotted to give the corresponding scales of paces or strides for any length pace or stride.

In case a field sketch is made in the scale of 1 inch to 800 feet or 1 inch to 96,000 inches (i.e., 1 inch on the map equals 96,000 inches on the ground) it would be found to be too small for detail, spotting of houses or other similar features, but since this sketch is made on cross-section paper it can easily be enlarged by the coordinate method to twice, four times, and eight times its size, as the case may be, to reproduce a sufficiently large map. In Figure 1, page 185, is shown the method of enlargement by the coordinate method. The original field sketch is on the scale of 1 inch to 800 feet plotted on cross-section paper. It is desired to enlarge this sketch to twice its size, or to the scale of 1 inch to 400 feet. This is easily done by doubling the coordinates as represented in the figure.

## 1. MATERIALS NECESSARY FOR MAP SKETCHING

### a. THE BOARD

A perfectly smooth soft pine board, preferably 14 inches by 13 inches, with absolutely squared edges is necessary, to which must be attached a small compass needle. A small watch compass is excellent for this purpose and may be affixed to the board by cutting a hole the size of the compass in order to hold it in a fixed position at all times. Place the compass in such a position as to have the north and south line parallel to one edge of the board. (See Figure 2, page 186).

### b. THE ALIDADE

The alidade is simply a small triangular piece of hard wood weighted with lead inserted in a small hole in the end to give it weight. It should be about 6 to 8 inches in length and have smooth surfaces on which to mark the various scales. This triangular scale or alidade is used as a sighting instrument and rests on top of the board. (See Figures 2 and 3, page 186).

### c. PAPER, PENCIL, AND THUMBTACKS

Any good grade cross-section or profile paper may be used, preferably ruled in one inch squares and divided into tenths. A pencil of hardness 4H is the best for field use. Thumbtacks are necessary to hold the paper in place on the board.

## 2. PREPARING THE SCALE FOR USE ON THE ALIDADE

As stated above, the length of the pace determined was 35.3 inches.

Therefore, placing the alidade along the line on the Chart P corresponding to 35.3 inches, transfer the divisions of this line to one edge of the alidade. (In this case see the dotted line on Chart P, page 183). This scale then will represent a scale in paces equal to 35.3 inches to correspond to the map scale of 1 inch to 800 feet. If the stride scale is preferred and it is believed the scale of strides will be found more convenient, the dotted line on Chart S, page 184, can be used, which is 70.6 inches as figures above. On the other edge of the alidade, directly opposite this pace or stride scale, lay off another scale in inches, this latter to be used in reading and scaling the map in the office.

### 3. METHOD OF SKETCHING THE MAP IN THE FIELD (FIAT SKETCH)

Assuming the sketching board is prepared with cross-section paper and compass, select a suitable point from which to start the sketch, preferably at the intersection of two streets on the extreme edge of the area to be mapped. With this as a starting point hold the board in such a position as to have one edge in a line parallel to the north and south line as shown by the compass needle. Now draw a line on the edge of the map in the north and south direction, marking the north end N. In all future positions when taking sights and readings be sure that the board always points in the same direction, namely north and south, parallel to the compass needles. This placing the board in the N and S line at each new position is known as orientation.

Figure 4, page 186, represents a portion of a town which it is desired to map, and shows intersecting streets A,B,C,D, and E. Since point A is on the extreme edge of town, assume this as the starting point. Holding the board in the hands, take position at A, orient the board until the N and S line on the map points north as indicated by the compass needle, then mark (a) on the map. Now (a) on the map corresponds to A on the ground. Facing the direction of B, place the alidade with one edge along point (a) on the sketch and sight along top edge to B, drawing a line in the direction of B indefinite in length. Without changing the position of the board, do the same for point D. Now from point A on the ground pace off the distance to B and with the pace scale on the alidade plot this distance on the sketch. Now with point (b) on the board over B on the ground, orient the board and sight to point C and in all other directions if at a street intersection, drawing the lines of directions on the sketch. After pacing to C and plotting the distance on the sketch, orient at C and repeat for points D and E. In this manner the sketch progresses from point to point with the board always in the same position with respect to the N and S line. The distance from point D to point A is the closing line and acts as a check on the accuracy of the sketch. At the same time that these distances are being paced off all important features along the streets may be noted by plotting the number of paces along the line and sighting at right angles to the line, recording the distance in paces to the object. For example: In Figure 4, midway between points A and B is a pond,, point A'. In pacing from A to B when opposite this pond at point A', plot the number of paces on the sketch, orient the board and sight to the pond, pace and plot as be-

fore. In this manner houses and other important features may be plotted on the sketch as it progresses.

Figure 5, page 187, is a sketch map made in this manner, the starting point being at point (a) and the closing point at (a'), the direction of the course taken being represented by the small arrows in the streets. The closing distance or difference is very small, considering the errors that might be present in inaccurate pacing, etc.

It is to be noted that sketch maps of this nature are plotted in the position of north and south as given by the compass needle which is not the true north, the north of the compass being known as the magnetic north. Therefore, this should be noted on the map.

#### 4. METHOD OF LOCATION BY INTERSECTION

In case of the inaccessability of points necessary of location, or to lessen the number of position "set-ups" the method of intersection may be found useful.

In Figure 6, page 187, the board is oriented at point I and sights taken at points, A,B,C, and D, and the lines of direction drawn on the map, marking the a',b',c', and d' as reference points.

Now when the board is progressed to point II and oriented, sights are again taken on points A,B,C, and D, and the lines of directions drawn on the map. The intersections of these lines with those marked a',b',c', and d' locate on the map the relative positions of points A,B,C, and D at the actual map scale.

In case of small areas, maps may be made entirely by making two or three well-chosen "set-ups" which command a view of all the principal features to be mapped; such a procedure is known as "position sketching".

The foregoing is the method of preparing flat sketch maps without regard to differences in elevation and other topographical features. Contours or imaginary lines of constant elevation are not necessary in the ordinary spot maps used in public health work, except in the case of malaria studies where the differences in elevation are vitally important for the purpose of determining drainage courses. Such maps for malaria control should, therefore, be prepared by an engineer with accurate instruments. For those who desire to acquaint themselves with the methods of plotting contours it is suggested that they obtain the little book published by the United States Infantry Association: Military Sketching and Map Reading by Grieves. This publication was used as a reference in preparing the present paper.

(Note: As this article was originally printed in the American Journal of Public Health, December, 1922, the pace and stride charts were rendered

useless by being reproduced on a smaller scale).

7665

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MAKING A PACE SCALE

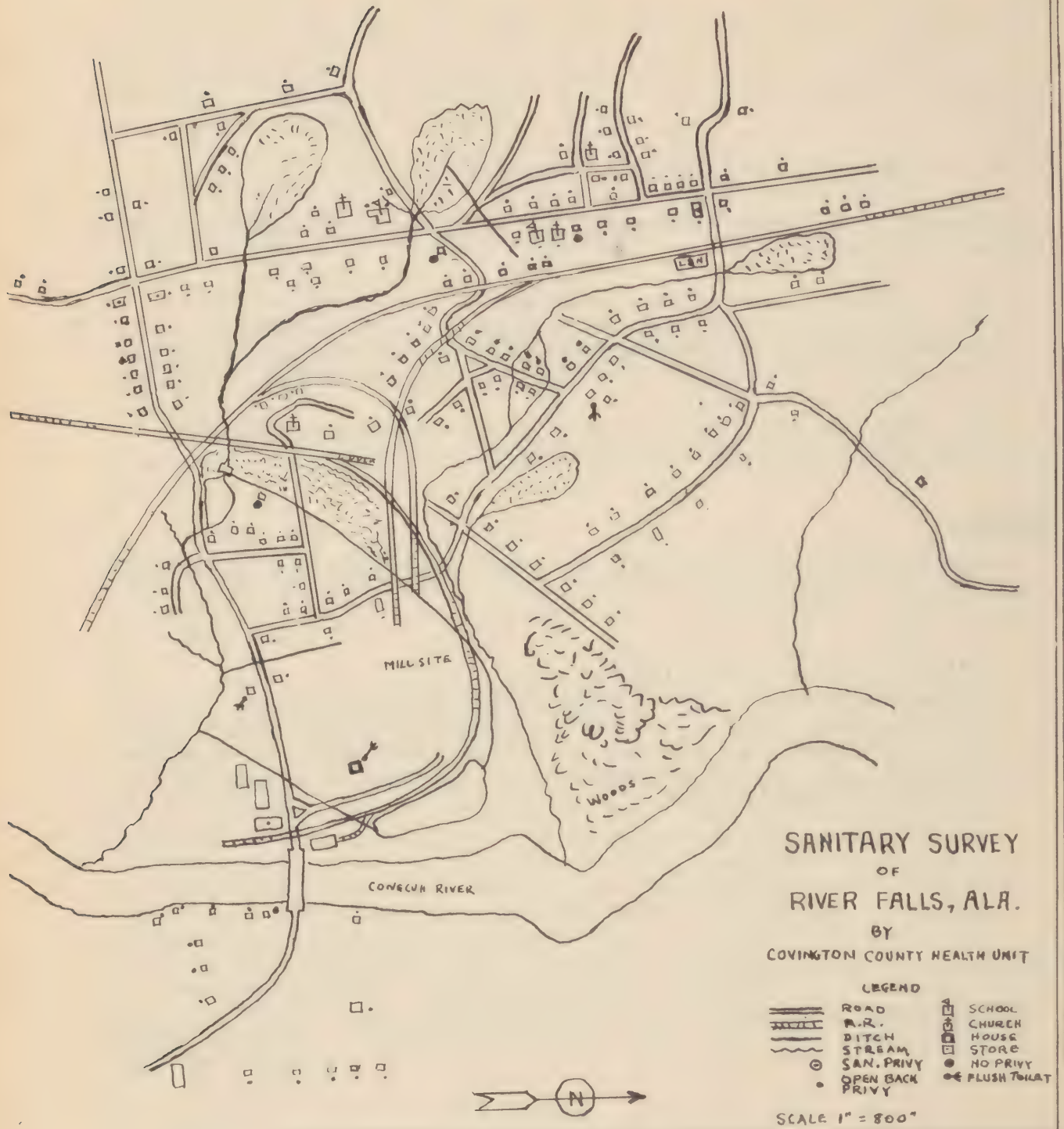
To make a pace scale for a map of any given scale, the following procedure is indicated:

1. Determine length of pace in inches.
2. Determine how many paces the pace scale should include.
3. Determine the RF (Representative Fraction) of the map (in inches).
4. Multiply (1) by (2) by (3), thus obtaining the length, in inches, of the pace scale..
5. Subdivide this pace scale into convenient pace groupings, and transfer to alidade, or any straight ruling edge.

EXAMPLE:

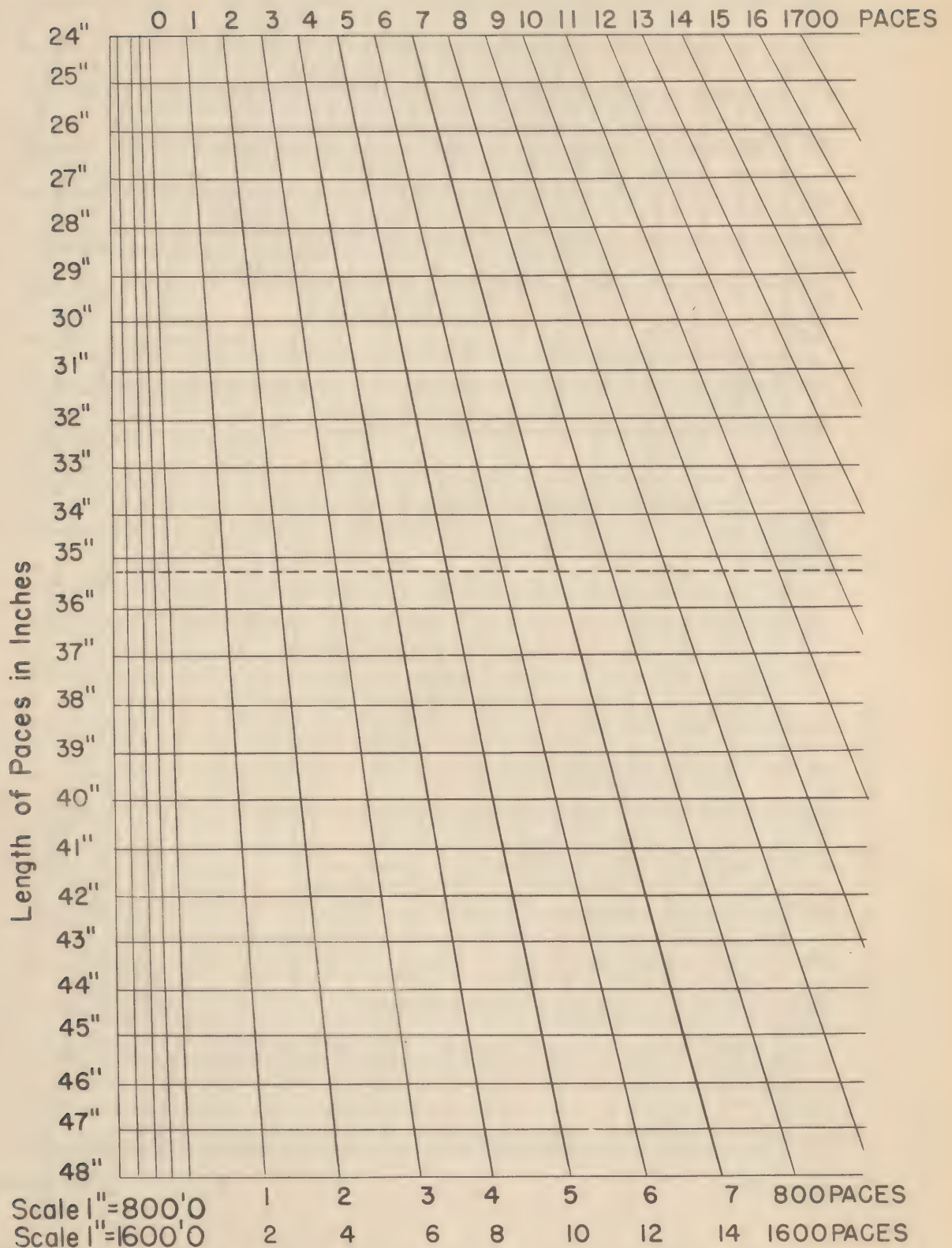
Problem: To make a pace scale for a map whose scale is 1 inch = 150 feet.

1. Length of pace = 32 inches.
2. Will make pace scale for 150 paces.
3. Map scale is 1" = 150' = 1800"; RF = 1/1800.
4.  $32 \times 150 \times 1/1800 = 2.67$  inches. (Equals 150 paces on map).
5. To subdivide the pace scale into 25 pace intervals, divide a line 2.67 inches long into 6 equal parts, and transfer to alidade, or edge of notebook, etc.



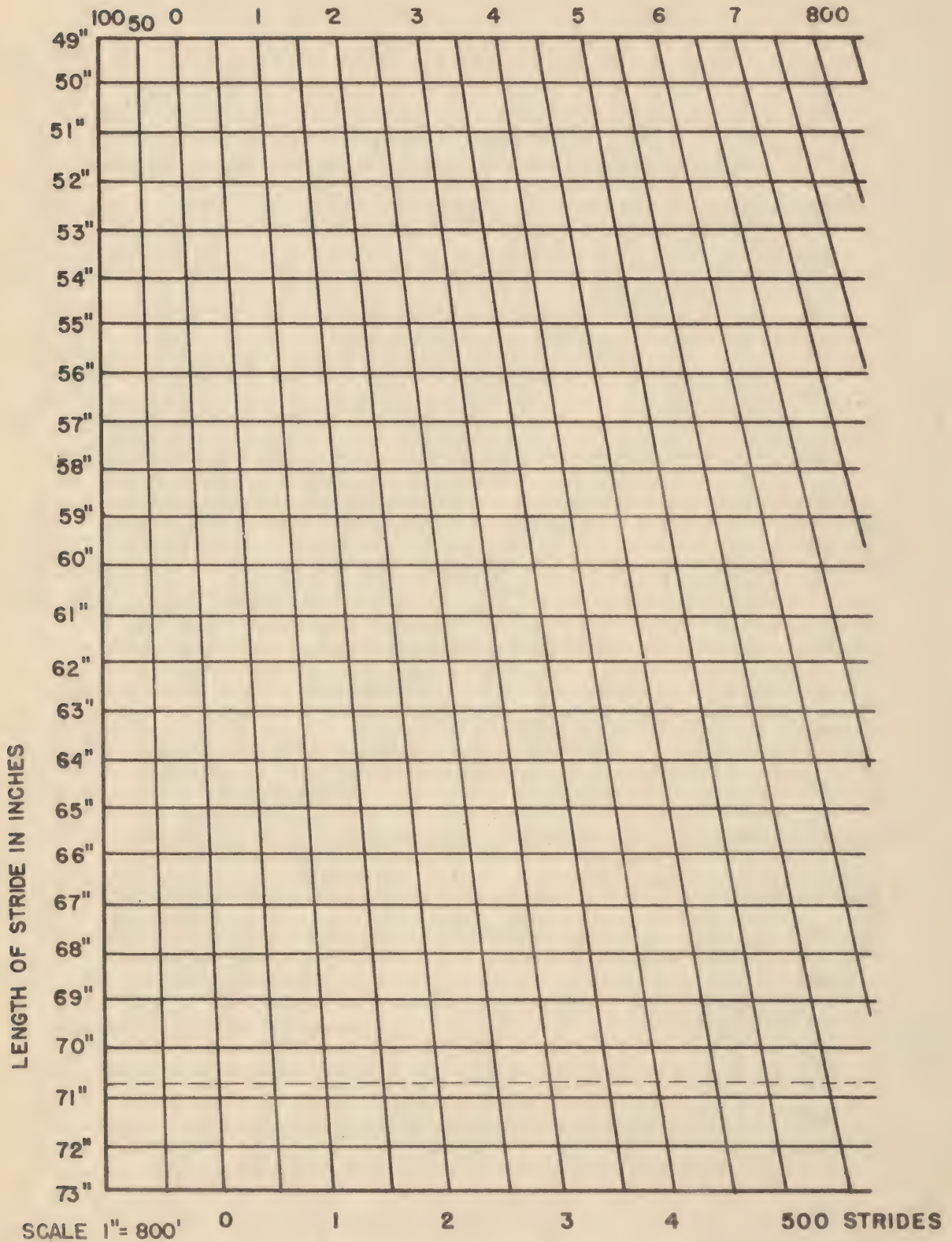
# CHART P. SCALE OF PACES

Scale: 1 inch = 9600 inches or 1 inch = 800'



# CHART S. SCALE OF STRIDES

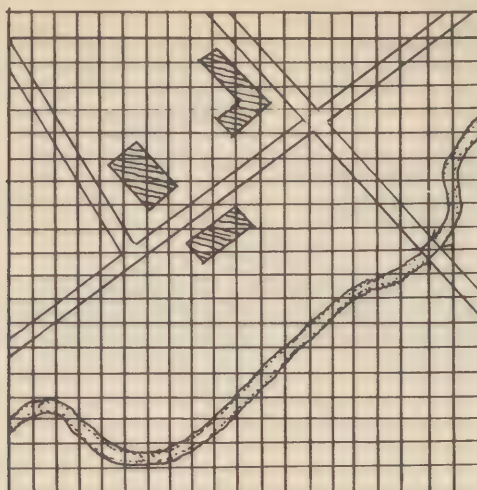
SCALE: 1 INCH = 9600 INCHES OR 1 INCH = 800 FEET



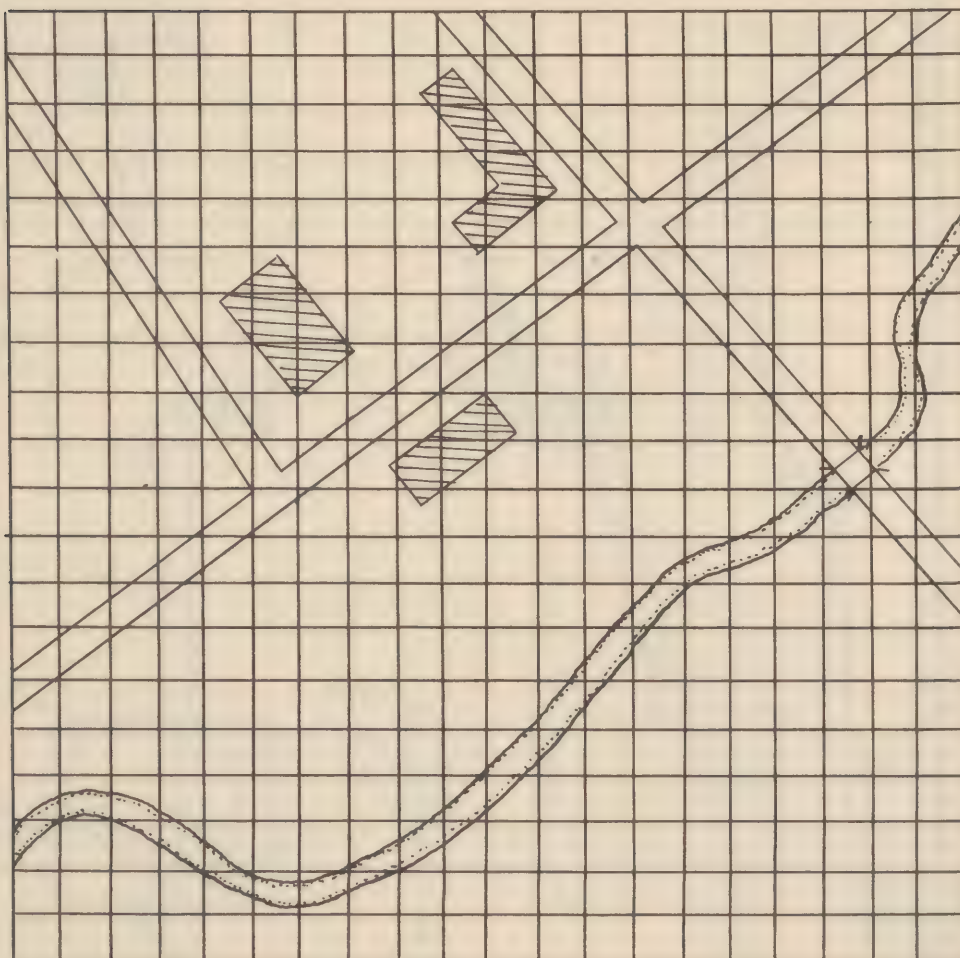
SCALE 1" = 1600' 1 2 3 4 5 6 7 8 9 1000 STRIDES

# METHOD OF ENLARGING BY COORDINATES

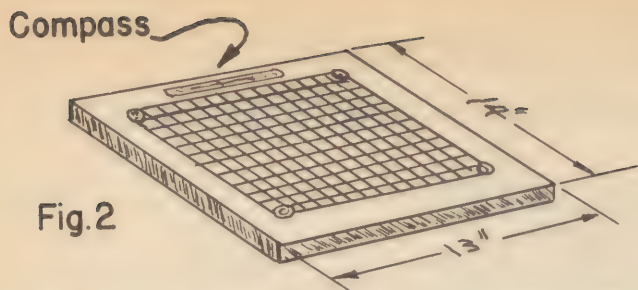
FIELD SKETCH of town on cross section paper Scale 1"=800'



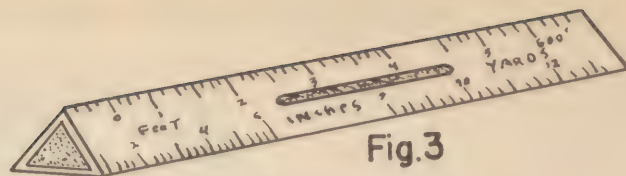
FIGURES I



FIELD SKETCH same as above enlarged to two times Scale 1"=400'

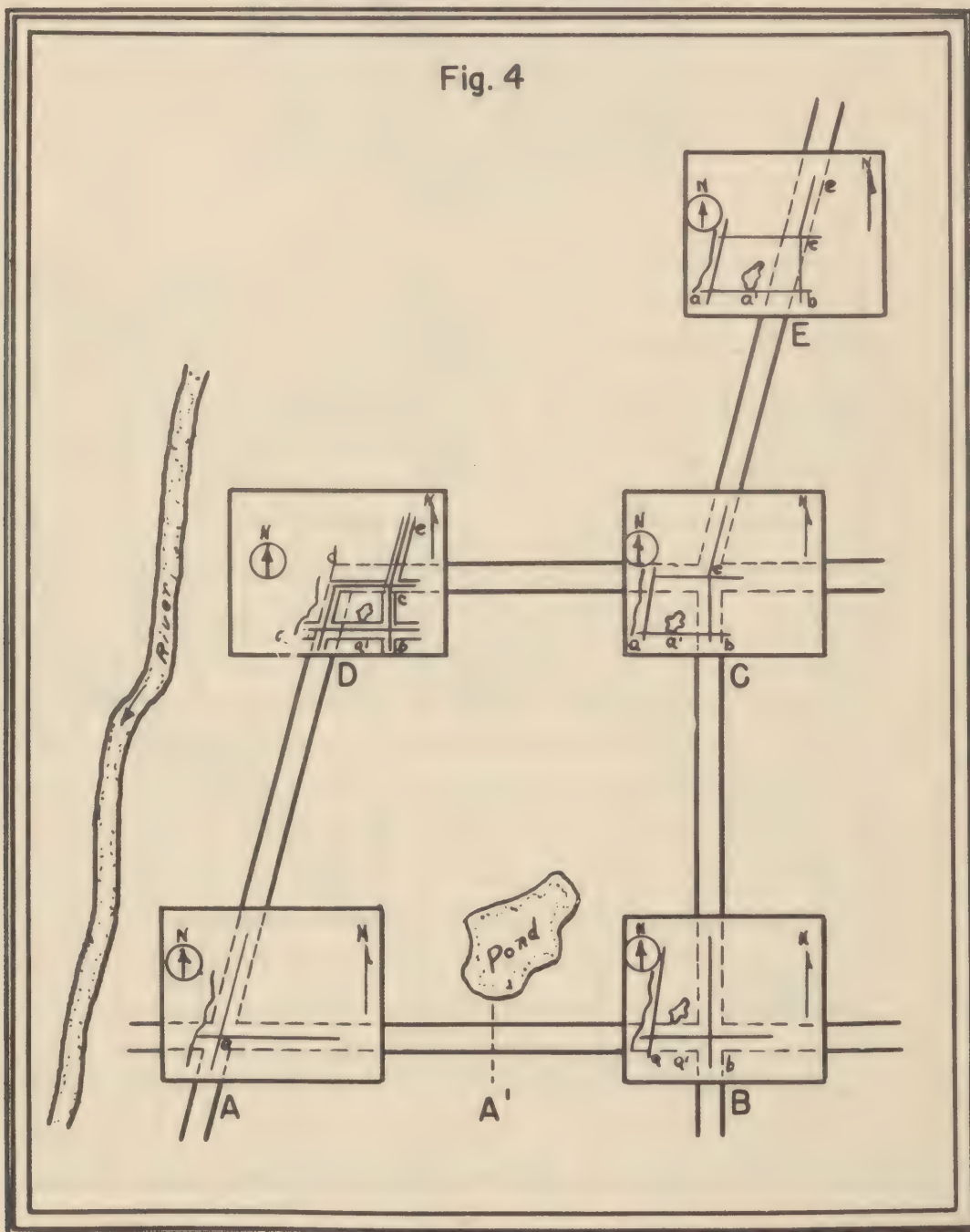


Sketching board with inset compass and cross-section paper in position

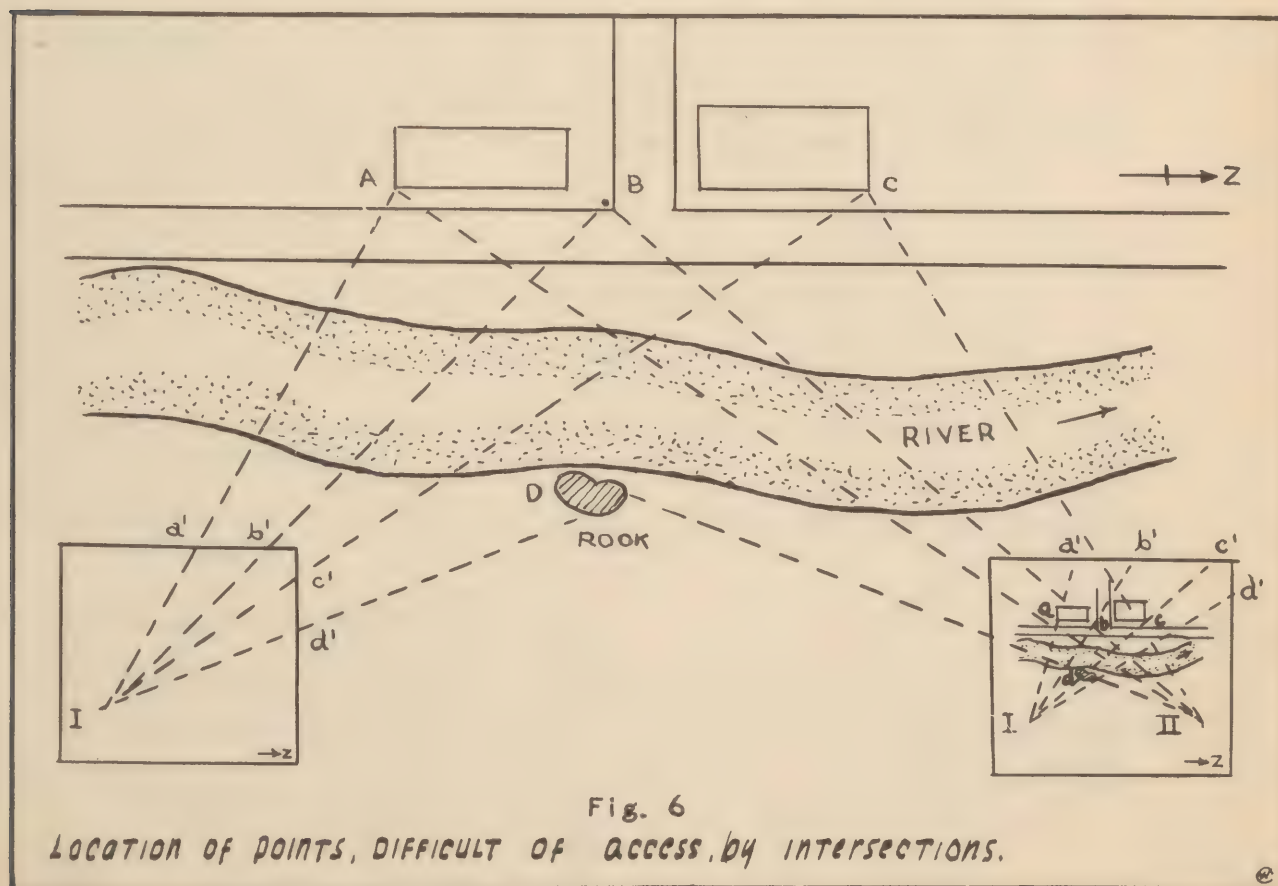
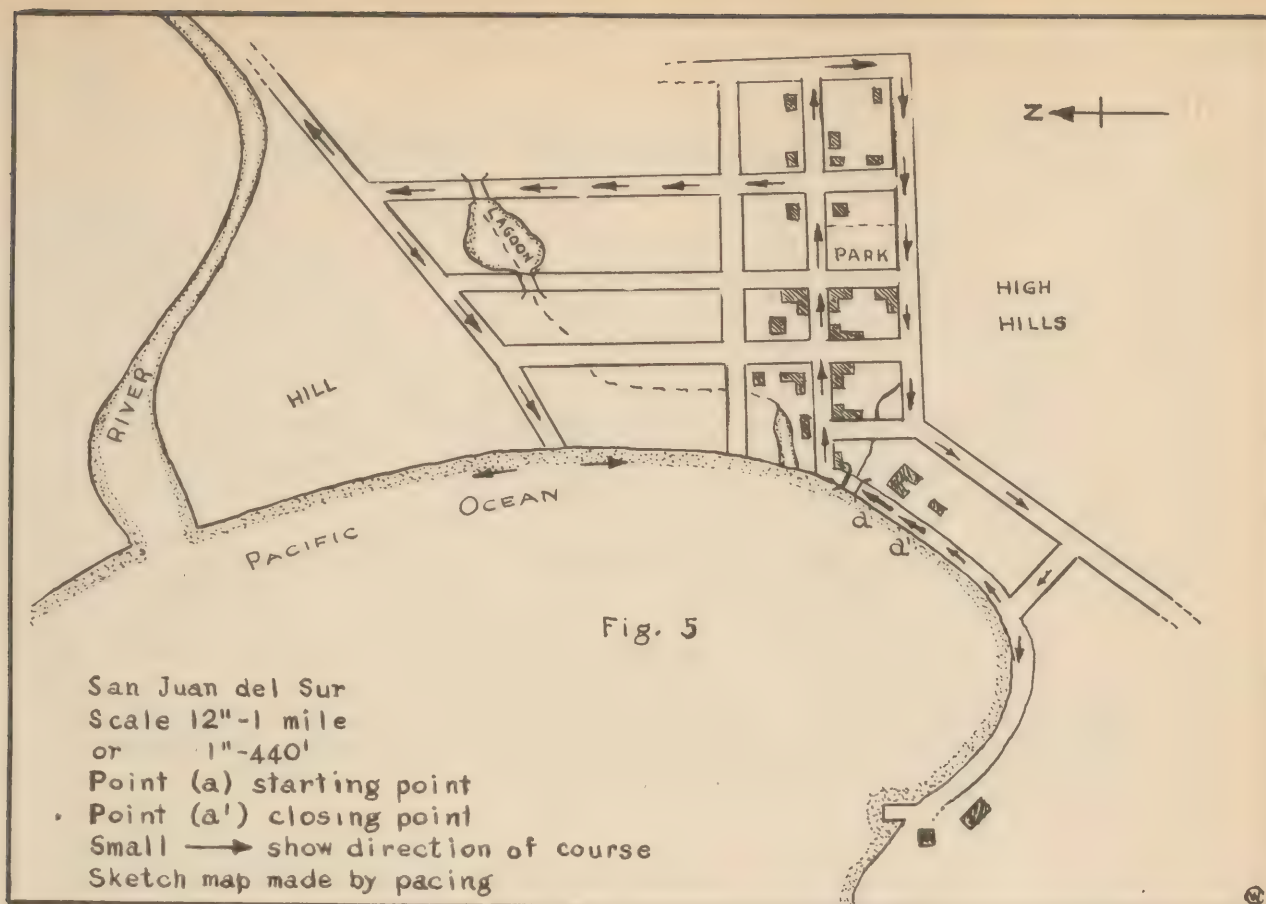


Combination triangular scale and alidade

### Flat Sketch of Streets



METHOD of ORIENTATION and PROGRESSION





## DRY METHOD PARIS GREEN MIXING APPLICATION

### 1. EQUIPMENT NEEDED

a. Paris green.

b. Type of dust used:

- |                 |              |                    |
|-----------------|--------------|--------------------|
| (1) Road dust   | (4) Charcoal | (7) Ashes          |
| (2) Fine gravel | (5) Talcum   | (8) Slacked lime   |
| (3) Sand        | (6) Silica   | (9) Fuller's earth |

Dust must be dry, so that Paris green will not adhere to it when it submerges.

c. Measures for measuring Paris green and diluent.

Advisable to have exact measures, namely, a 25 c.c. cup for Paris green and a 500 c.c. container for dust if a 5% mixture is used; other appropriately sized containers for different percents of mixture.

d. Mixing machine (included in appendix).

e. Knapsack or other type duster complete with hose and nozzle.

### 2. PROCEDURE

a. Select finest local dust procurable. (If charcoal is available, it is an excellent diluent, but must be ground). Always have a good supply on hand.

b. Pass dust through screen.

c. Place required amount of dust in mixing machine.

d. Add required amount of Paris green, 1% to 5%.

e. Revolve mixer at least 200 times, using a slow-start-stop motion (about 6 minutes).

f. Remove dust from mixer and place in gunny sack for transportation to field.

### 3. IN THE FIELD (Knapsack Sprayer)

a. Fill knapsack sprayer with Paris green dust mixture.

b. Proceed to apply to water surface by operating knapsack sprayer

handle. (If clogging occurs, usually slight tapping will remove the difficulty).

- c. Proceed at a slow walk, pointing nozzle down toward water and about 10 inches above water, being sure that every square inch of water surface is covered.

#### 4. QUANTITY OF PARIS GREEN TO BE USED

0.1 c.c. (0.125 gr., 1.9 grains) to a square meter of surface.

This is equivalent to one litre (1,250 gr.) over 10,000 square meters of surface or approximately one pound per acre --After Hackett.

#### 5. OTHER METHODS OF APPLICATION

- a. By hand. The mixture is thrown by hand as in sowing grain. The mixture is carried in a bag suspended from the shoulder.
- b. By hand bellows. Highly unsatisfactory from an operational point of view.
- c. By rotary blowers. A very satisfactory method provided the blower is properly made. "Balance of Machine" should be considered and protection of gears is essential.
- d. Airplane dusting, when large areas are involved. Use of an airplane is an involved procedure, but can be most successful. Percentage of Paris green in mix must be greater when used for airplane dusting, not less than 10%, and frequently, with high wind velocities. much higher.

For other methods of application of Paris green, see SG Letter, Number 22, page 207.

## SLOPES, VELOCITIES, AND SOILS

### Side Slopes

Cemented gravel, stiff clay soils, ordinary hardpan	3/4 to 1*
Firm, gravelly, clay soil, or sidehill cross-sections in average loam	1 to 1
Average loam or gravelly loam	1 1/2 to 1
Loose, sandy loam	2 to 1
Very sandy loam	3 to 1
(*) 3/4 horizontal to 1 vertical	

### Maximum Mean Velocities Safe Against Erosion

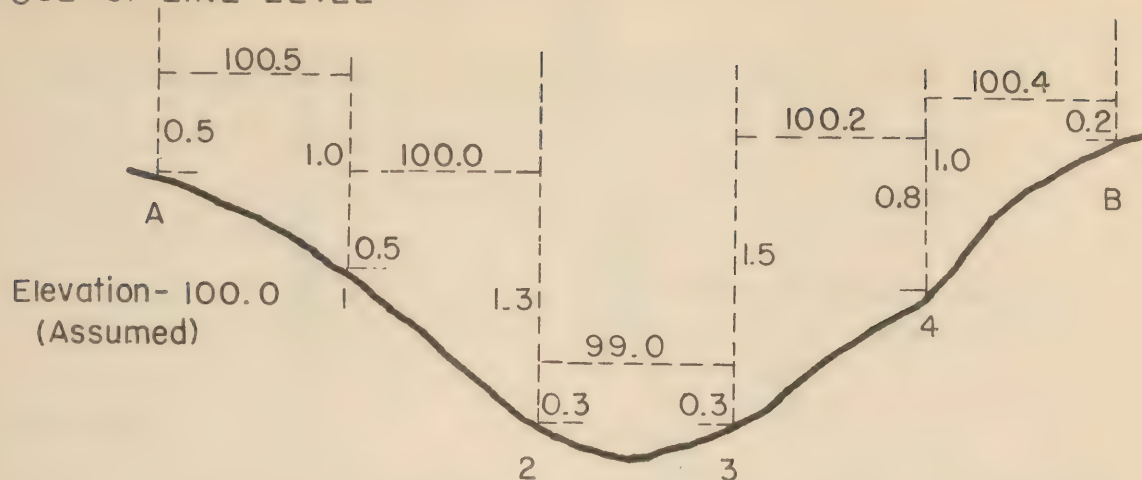
Nature of Canal Bed	Ft./Sec.
Very light pure sand of quick-sand character	0.75 - 1.00
Very light loose sand	1.00 - 1.50
Coarse sand or light sandy soil	1.50 - 2.00
Average sandy soil	2.00 - 2.50
Sandy loam	2.50 - 2.75
Average loam, alluvial soil	2.75 - 3.00
Firm loam, clay loam	3.00 - 3.75
Stiff clay soil, ordinary gravel soil	4.00 - 5.00
Coarse gravel, cobbles, shingles	5.00 - 6.00
Conglomerates, cemented gravel, soft slate, tough hardpan, soft sedimentary rock	6.00 - 8.00
Hard rock	10.00 - 15.00
Concrete	15.00 - 20.00

### Classification of Soils

1. Alluvium is the finer deposit of earth, sand, gravel, and other transported material which has been washed away and deposited by rivers, floods, or other causes on land and not permanently submerged.
2. Clay is the general name for cohesive soils, which are firmly coherent, weighty, compact, and hard when dry, but stiff, viscid, and ductile when moist, and smooth to the touch. Clay absorbs water greedily, but not readily, is diffusible in water, and, when mixed, does not readily subside in it.
3. Gravel consists of small stones or fragments of stone or very small pebbles larger than particles of sand, but often mixed with them.

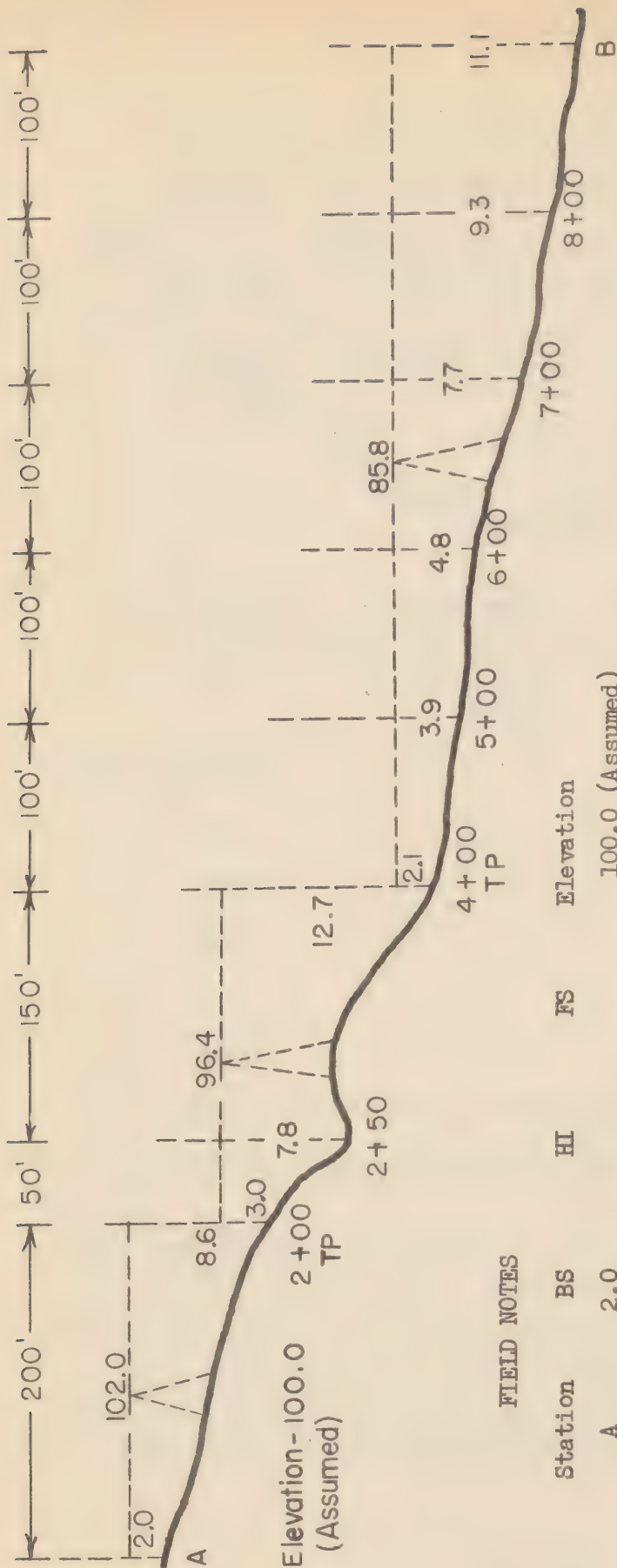
4. Hardpan is a rather loosely used term, but is most commonly applied to a very dense heterogeneous mass of clay, sand, and gravel of glacial drift origin; it is also applied to the hardstratum of consolidated soil underlying the surface soil.
5. Loam is a mixture of sand and clay with oxide of iron, and generally a varying amount of organic matter:
6. Sand is any mass or collection of fine particles of stone, particularly of siliceous stone, but not strictly reduced to powder or dust.

# USE OF LINE LEVEL



(a) Station	(b) Distance of String Above Ground at Sta. of Known Elev. (Backsight)	(c) Level of String Above Reference Plane (HI)	(d) Distance From String to Ground at Sta. Whose Elevation is Sought (Foresight)	(e) Elevation of Ground at Station
A	0.5	100.5		100.0 (Assumed)
1	0.5	100.5	1.0	99.5
2	0.3	99.0	1.3	98.7
3	1.5	100.2	0.3	98.7
4	1.0	100.4	0.8	99.4
B			0.2	100.2

- Procedure:
1. Assume ground elevation at A and enter in column (e).
  2. Tie end of string to stake at A.
  3. Measure distance from ground to string and enter in column (b).
  4. Add column (b) to ground elevation (col(e)) and enter on next lower line in column (c).
  5. Tie other end to stake at Sta. 1, and level by use of line level.
  6. On stake at Sta. 1 measure distance from string to ground. Enter in column (d) at Sta. 1.
  7. Subtract reading in column (d) from column (c) above, and enter in column (e). This is then the ground elevation at Sta. 1.
  8. Since the ground elevation at Sta. 1 is now known, we repeat the above procedure and determine the ground elevation at Sta. 2, and so on to Sta. B, thus determining the difference in elevation between A and B.



# FIELD NOTES

Station BS HI FS Elevation  
 A 2.0 102.0  
 2+00 TP 3.0 96.4 8.6 93.4  
 2+50 7.8 88.6  
 4+00 TP 2.1 85.8 12.7 83.7  
 5+00 3.9 81.9  
 6+00 4.8 81.0  
 7+00 7.7 78.1  
 8+00 9.3 76.5  
 B 11.1 74.7

## DIFFERENTIAL LEVELING

Difference in Elevation is 100.0 - 74.7 25.3 ft.

BM - Benchmark.

BS - Backsight. A sight at a point whose elevation is known.

FS - Foresight. A sight at a point whose elevation is sought.

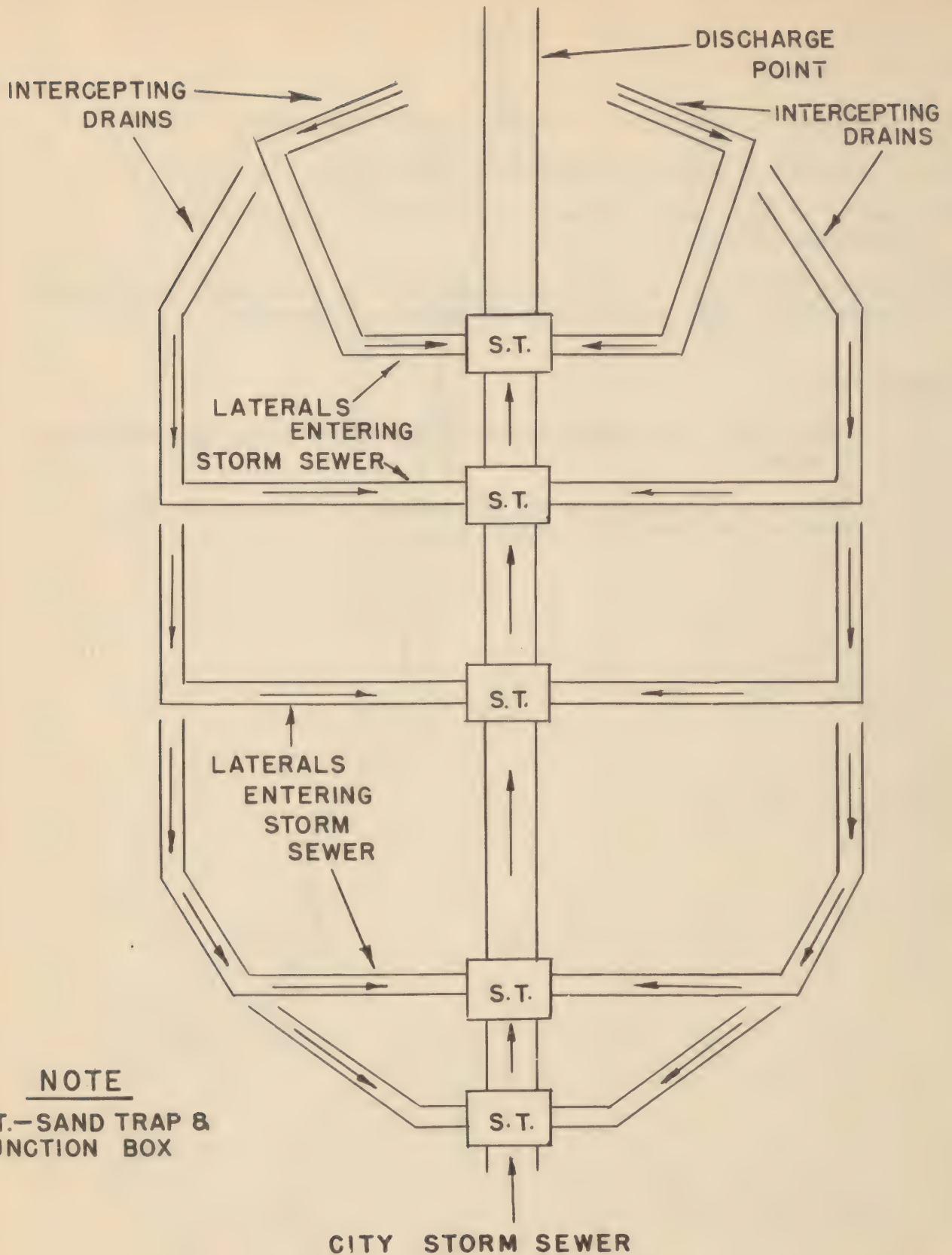
HI - Height of Instrument. Elevation of the line of sight above the reference plane.

TP - Turning Point. A station at which elevation is determined, and to which backsight is taken to establish new Height of Instrument.

### Precautions

1. Between TPs keep length of backsights and foresights approximately equal.
2. As soon as HI is determined, a foresight is taken on next TP; then intermediate points picked up.

APPROXIMATE — PLAN OF  
PENSACOLA — PROJECT



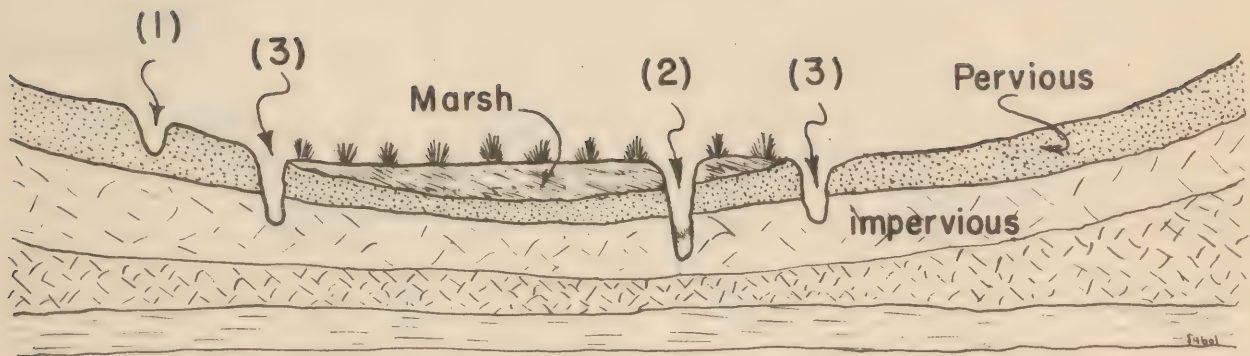
NOTE

S.T.—SAND TRAP &  
JUNCTION BOX

OBSERVATION REGARDING LOCATION OF INTERCEPTION  
DRAINAGE DITCHES WHEN DRAINING SEEPAGE AREA.

Poor Location.....No. 1 & 2

Good Location.....No. 3

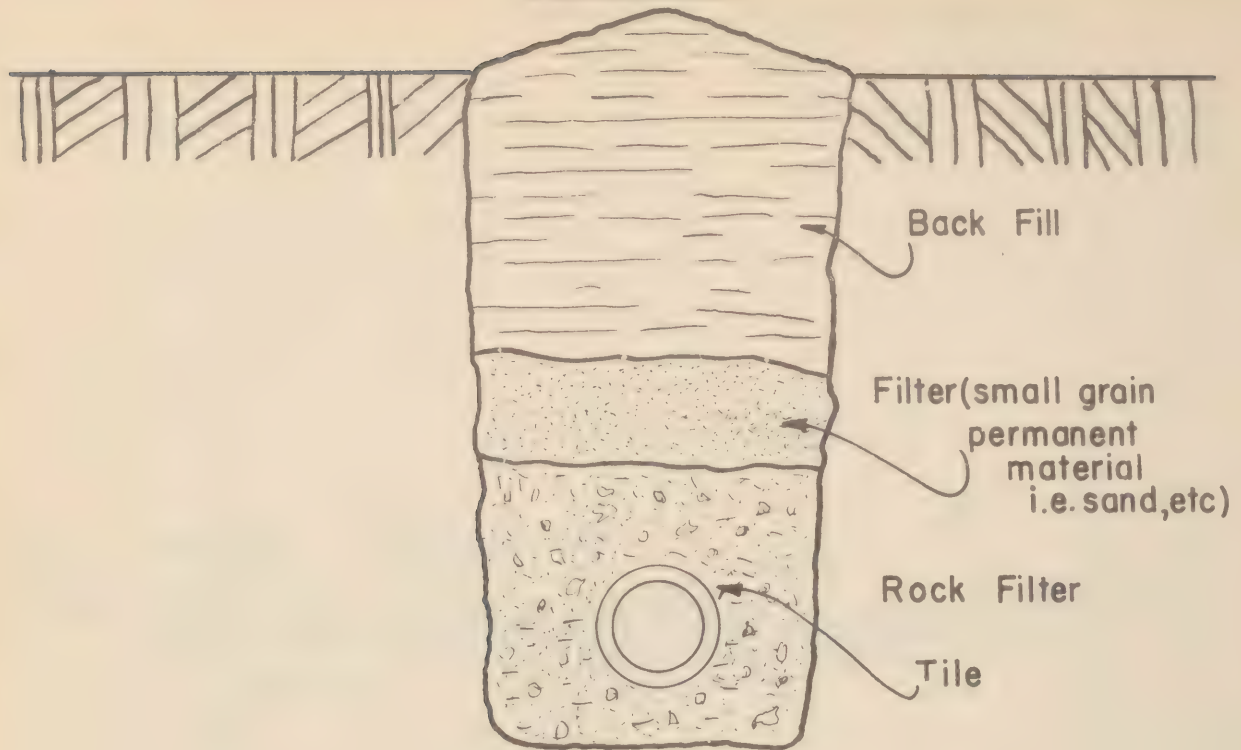


- (1) Canal insufficiently deep. Does not penetrate impervious layer. Seepage may flow under excavation.
- (2) Canal excavated inside edge of marsh, and needlessly deep. Probable that small area between canal and marsh edge would remain a menace.
- (3) Arrow "3" indicates better location of interceptor canal than at location "2".

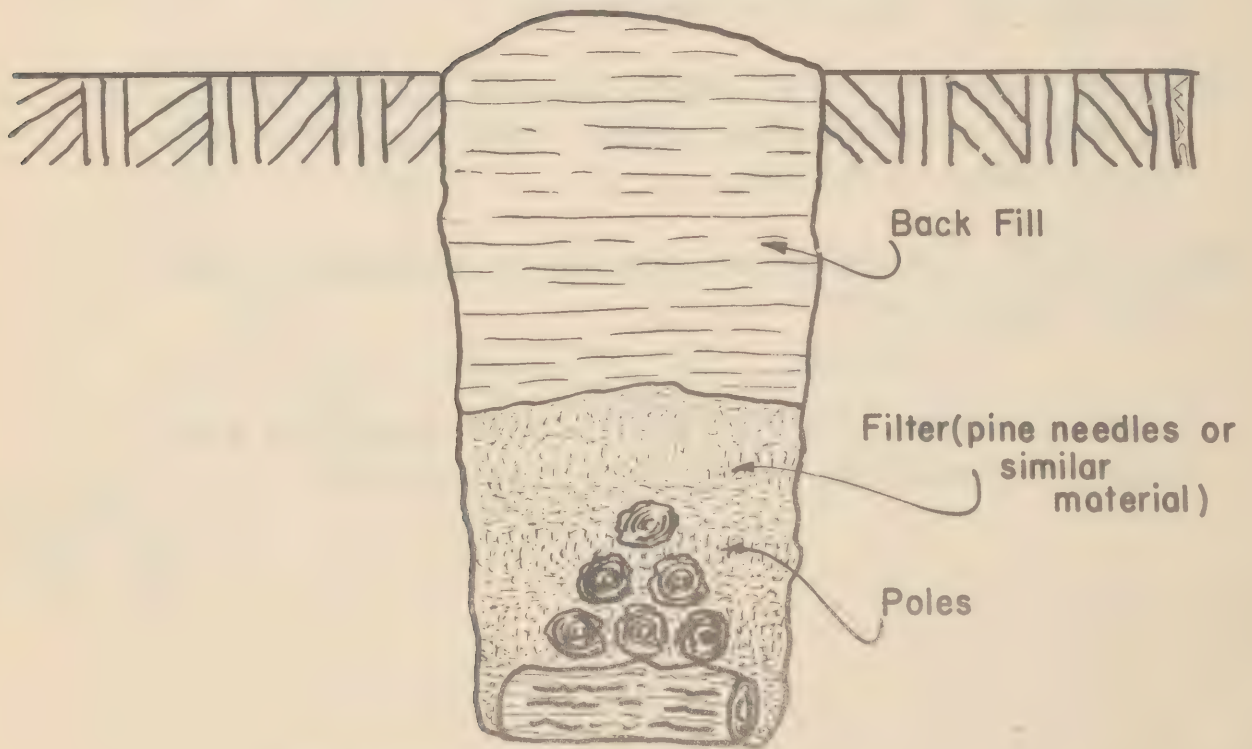
Under situation depicted above ditch should be dug of a depth to reach the "impervious" layer of soil.

# UNDERGROUND DRAINS

## Permanent

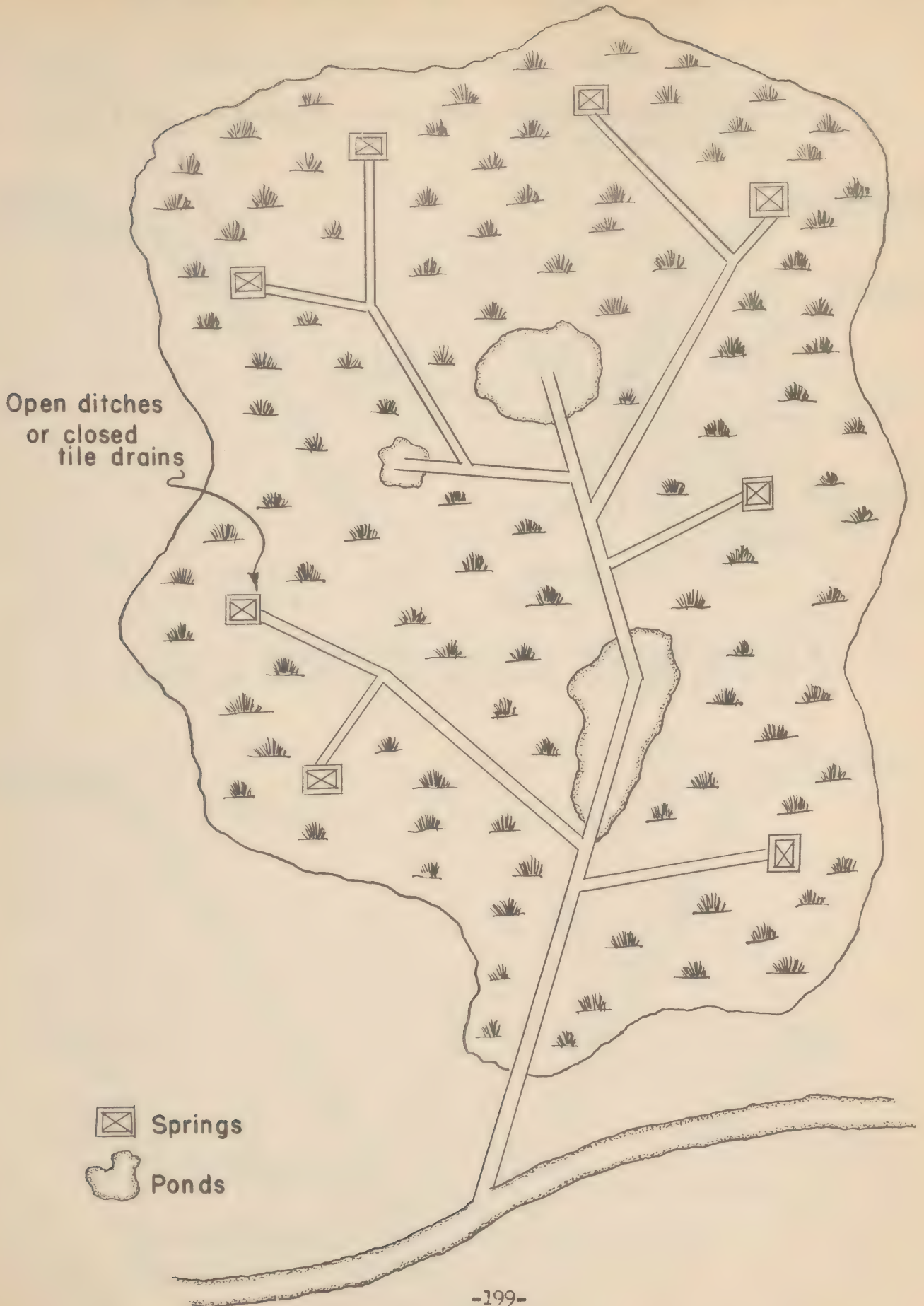


## Temporary



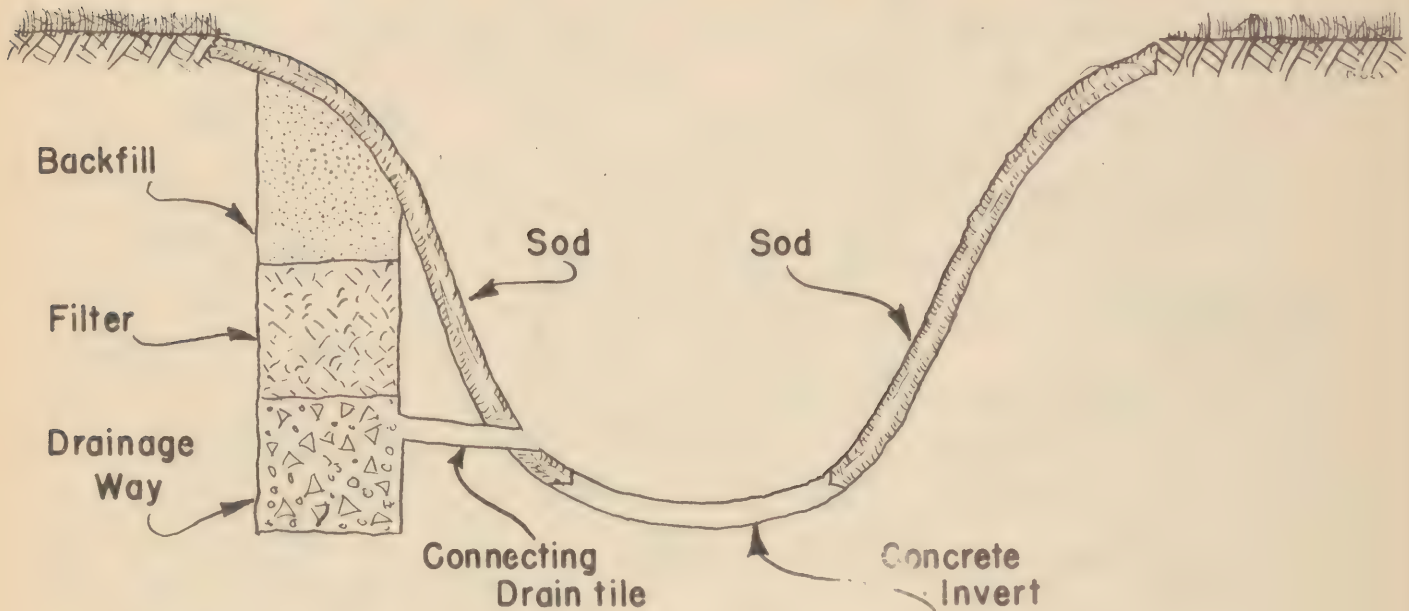
SO CALLED "POLE DRAIN"

# DRAINAGE OF SPRING FED MARSHES AND PONDS

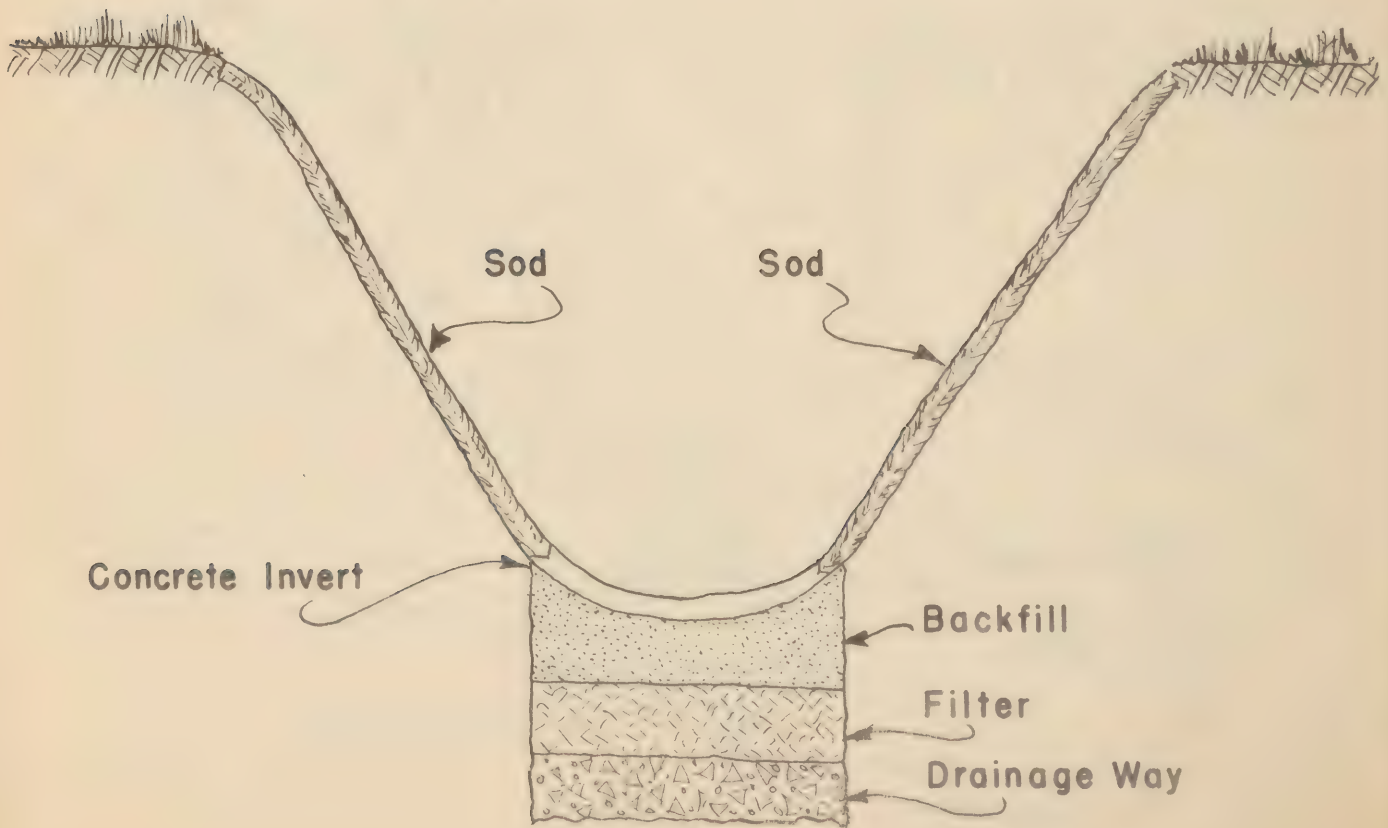


**CROSS SECTION OF PERMANENT DITCH  
WITH CONCRETE INVERTS AND SOD REVETMENTS**

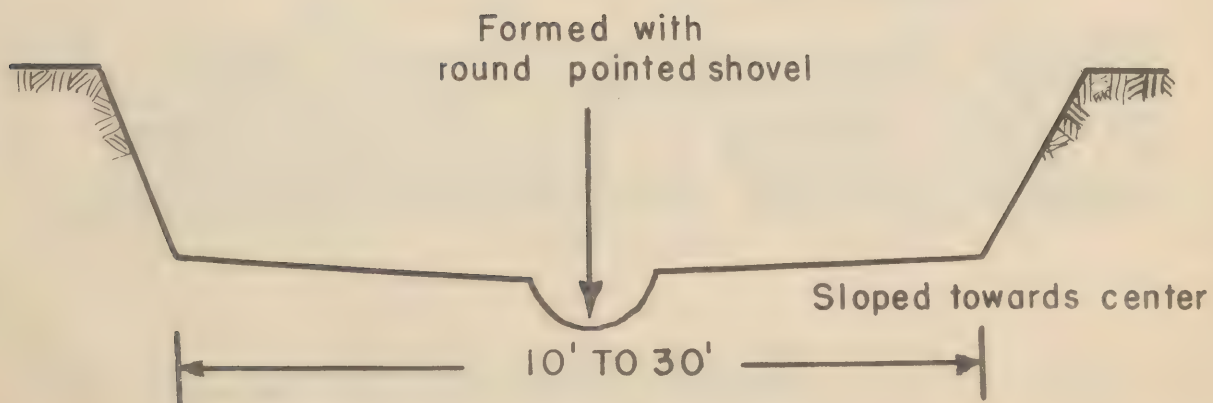
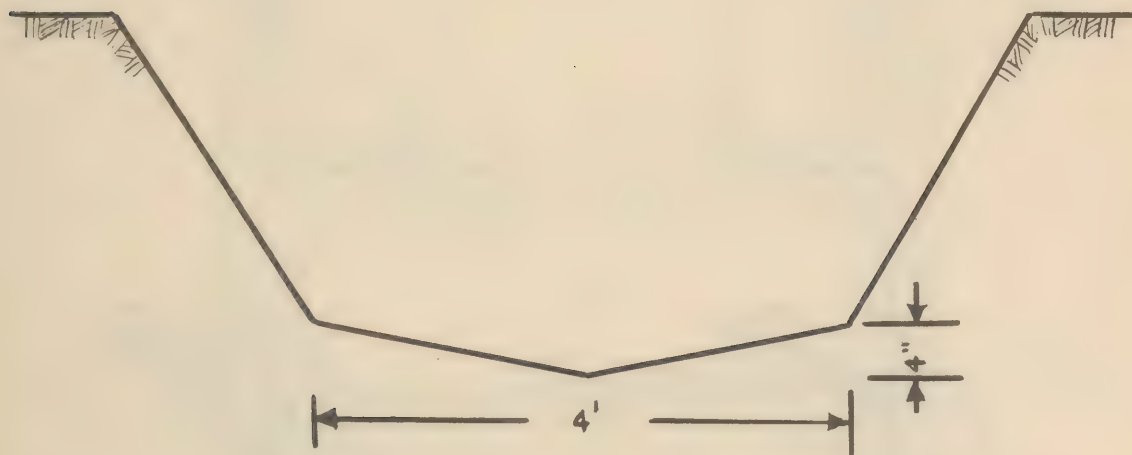
- A. For use in unstable soils or where hydraulic pressure is great on the side slope.  
Laterally placed straight sided ditches installed to relieve lateral hydraulic pressure.

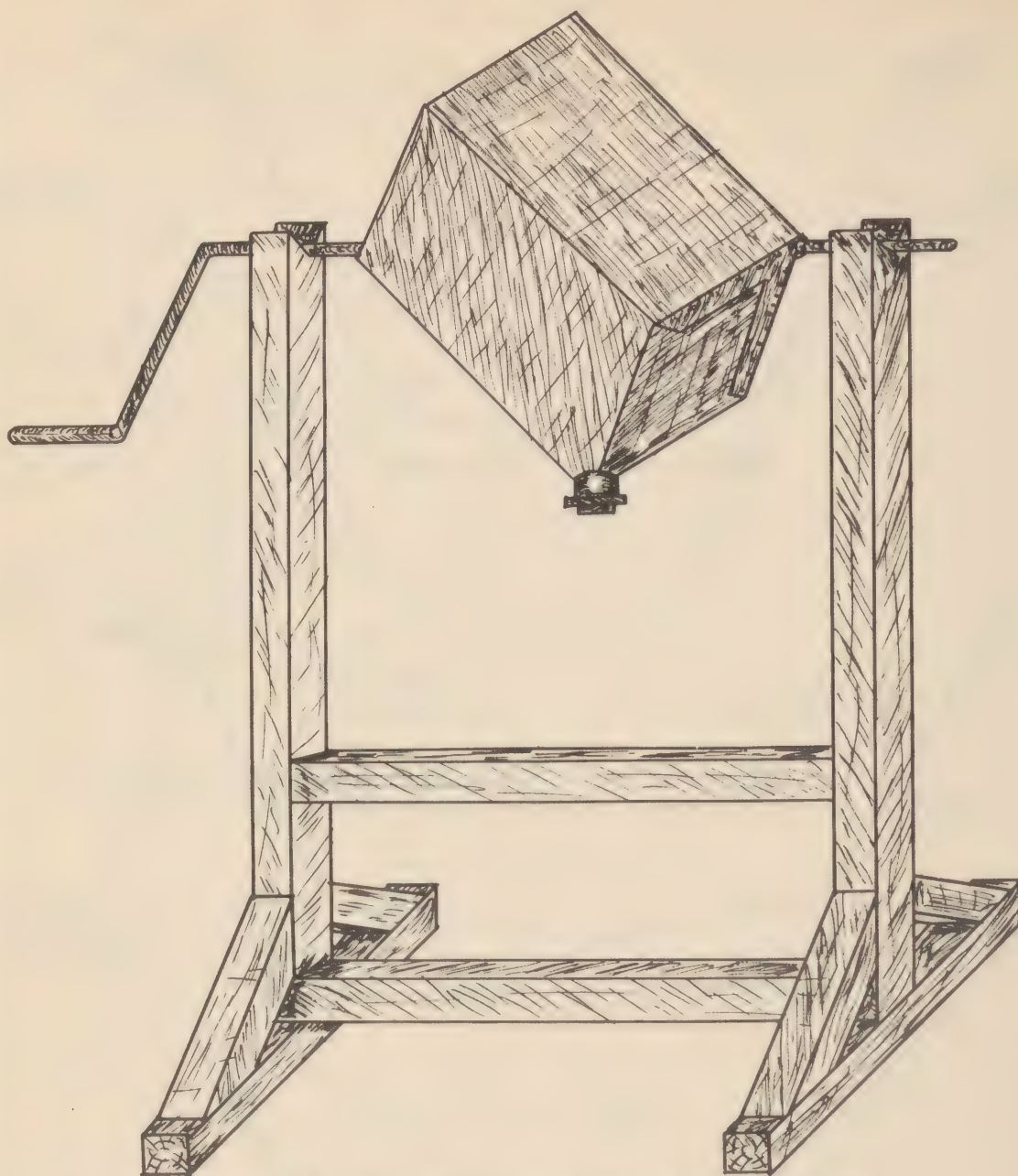


- B. For use where sub-grade requires drainage.  
Generally employed where residual pot-holes are formed.



# IDEAL DITCH CROSS-SECTIONS



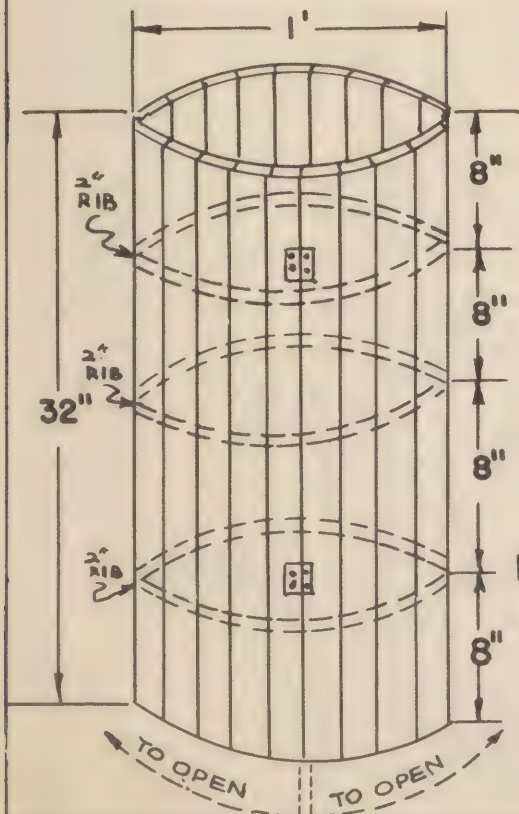
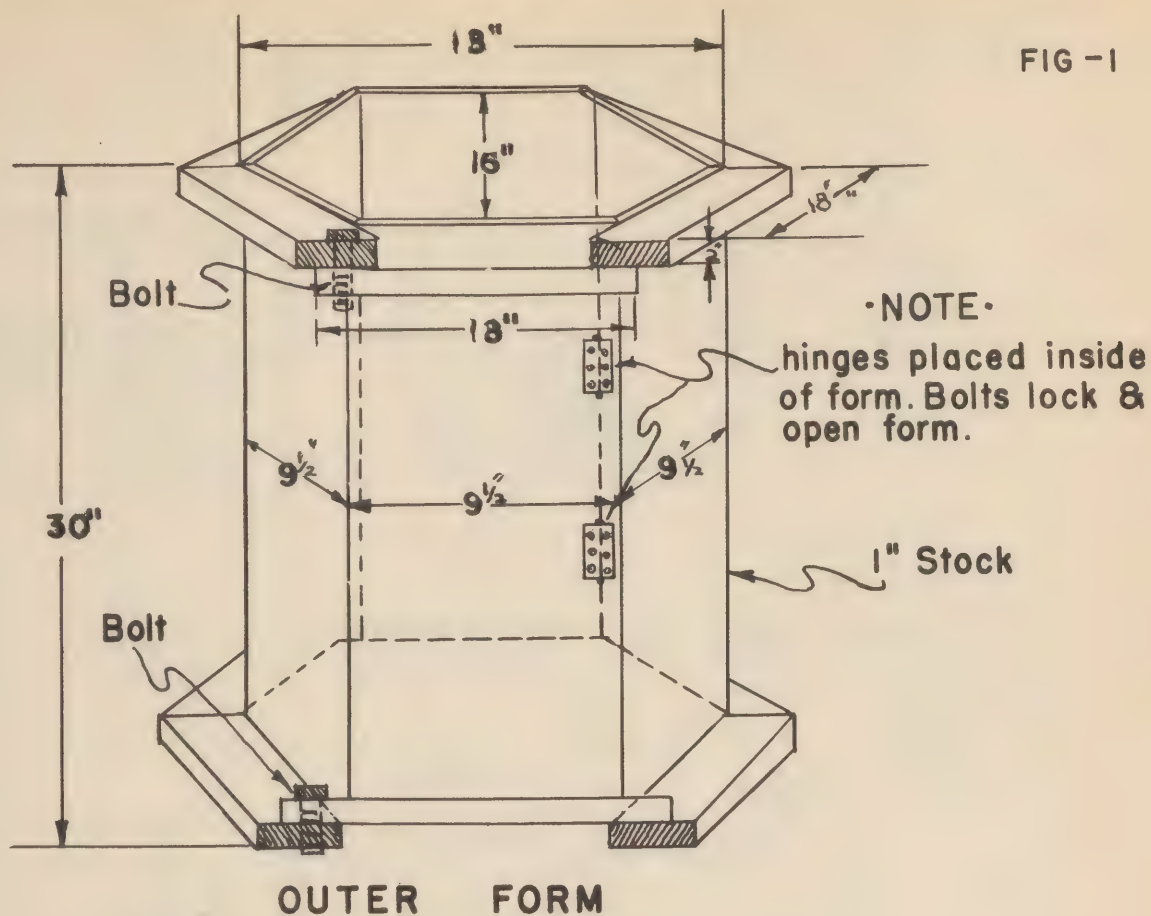


MIXER FOR USE WITH PARIS GREEN

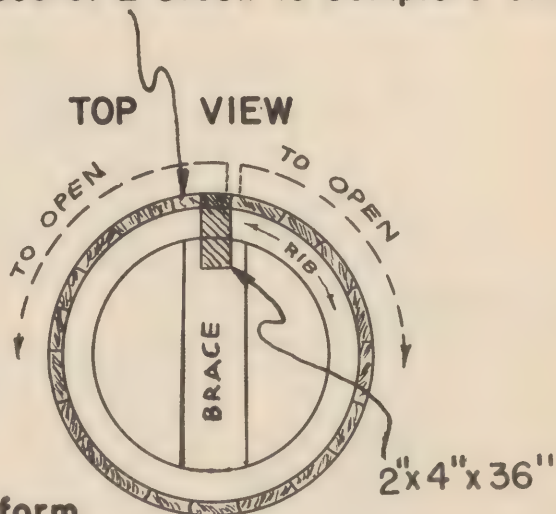
A SUITABLE APPARATUS MAY BE CONSTRUCTED FROM A RECTANGULAR BOX OF GALVANIZED IRON SUSPENDED ON TWO PILLARS FROM DIAGONALLY OPPOSITE CORNERS, AND TURNED BY A HANDLE. A CONVEIENT SIZE IS 33X48 CM. (ABOUT 13X19"). AROUND 200 TURNS ARE NECESSARY FOR SATISFACTORY MIXING

# CONCRETE PIPE FORM WOOD

FIG -1

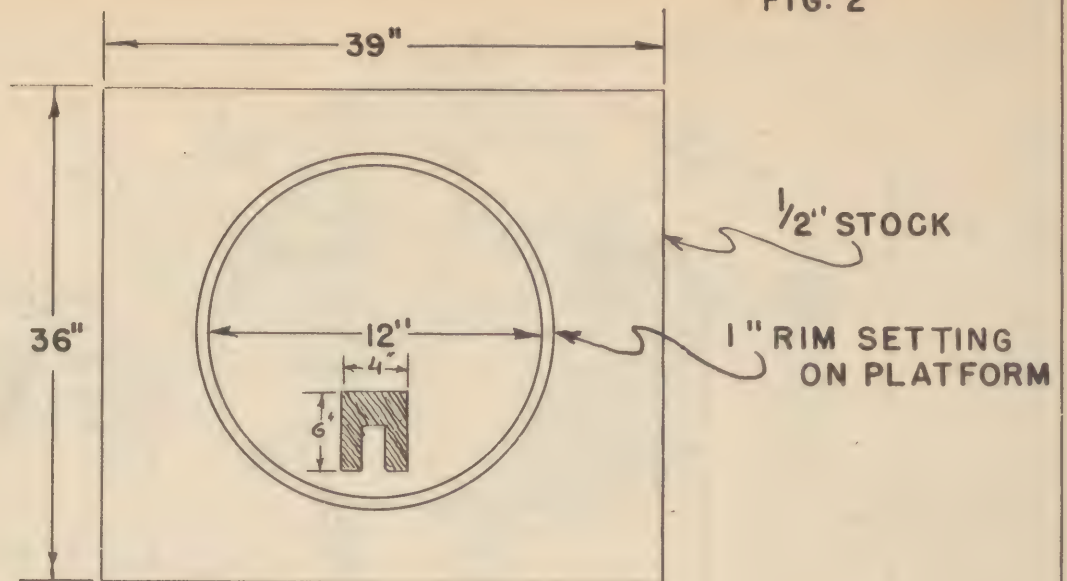


20 Pieces of 2" Stock to complete circle

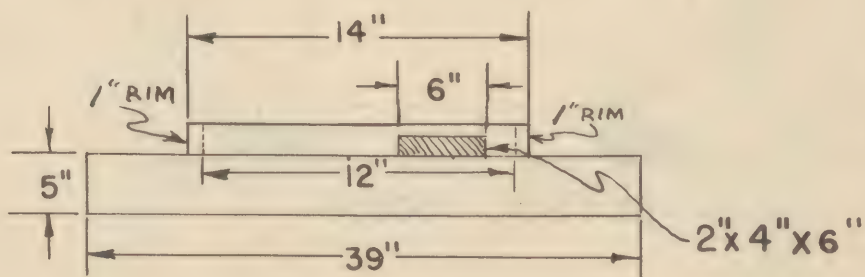


# CONCRETE PIPE FORM WOOD CONT.

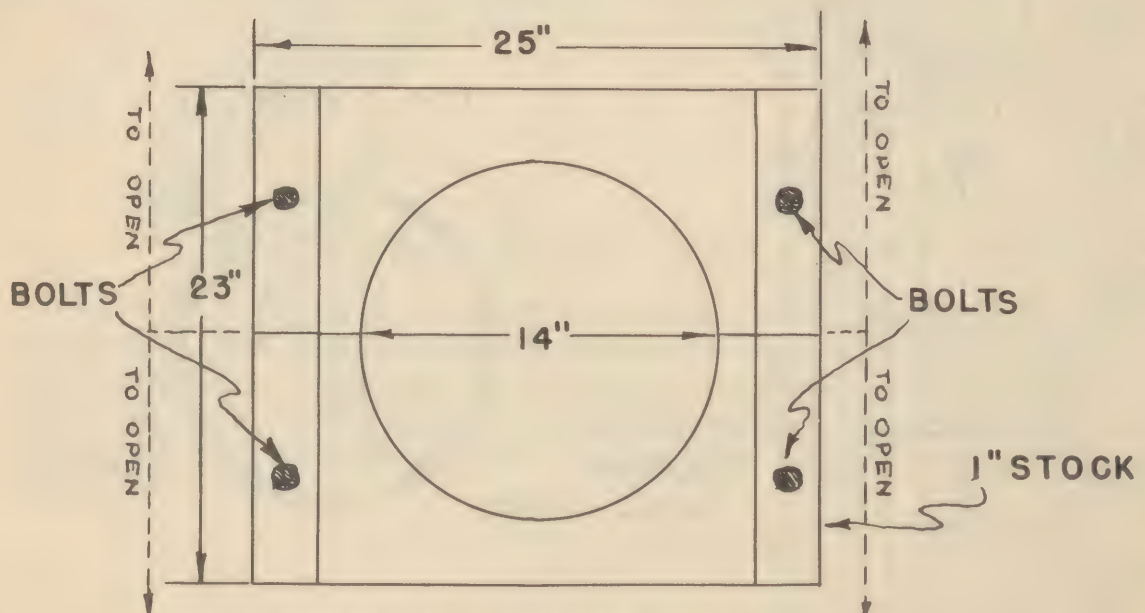
FIG. 2



TOP VIEW OF PLATFORM



SIDE VIEW OF PLATFORM



TOP VIEW OF WRINGER HEAD

**CONCRETE PLANT-ESCAMBIA COUNTY HEALTH DEPT.  
MATERIALS NECESSARY AND DETAILED COST PER UNIT PRODUCED**

PRODUCT	QUANTITY OF MATERIAL				COST OF MATERIAL		MATERIALS TOTAL COST	LABOR COST PER MAN HR. PER UNIT	25¢	TOTAL COST
	CEMENT	SAND	GRAVEL		CEMENT	SAND	GRAVEL			
24" STD. TILE 2" WALL	6.72 BAG	0.080 C.Y.	0.053 C.Y.		38.9¢	10¢	11.9¢	60.8¢	2.5	62.5
24" H.D. 5" WALL	1.4 BAG	0.155 C.Y.	0.103 C.Y.		75.6¢	19.4¢	23.3¢	118.3¢	3.2	80.0
18" STD. TILE 1 5/8 WALL	0.54 BAG	0.06 C.Y.	0.04 C.Y.		29¢	8¢	9¢	46.0¢	2.22	55.6
6" TILE SUBSOIL	0.033 BAG	.0036 C.Y.			1.8¢	0.5¢		2.3¢	300 TILE IN 16 MH	1.3
4" TILE SUBSOIL	0.02 BAG	.0022 C.Y.			1.1¢	0.3¢		1.4¢	500 TILE IN 16 MH	0.78
INVERT PANAMA	0.25 BAG	0.021 C.Y.	0.028 C.Y.		13.5¢	2.6¢	6.3¢	23.4¢*	.33	8.25
SIDE WALL PANAMA	.114 BAG	.01 C.Y.	.0126 C.Y.		6.2¢	1.3¢	2.8¢	10.3¢	.22	5.56
INVERT	0.24 BAG	0.0267 C.Y.	0.0177 C.Y.		13¢	3.3¢	4¢	20.3¢	.83	20.8
1/3 OF 24" TILE										41.1¢

**COST OF MATERIALS**

CEMENT \$2.15 BARREL (4 BAGS)  
SAND 1.25 CUBIC YARD  
GRAVEL 2.25 CUBIC YARD

**MIX**

INVERTS (PANAMA)  
SIDEWALLS  
18" X 24" TILE  
4" X 6" TILE

CEMENT SAND GRAVEL WATER (\*\*)  
1 2 1/4 3 6 GALS.  
1 2 1/4 3 6 GALS.  
1 3 2 1 1/2 GALS.  
1 3 0 2 GALS.

(\*) INCLUDES 1¢ FOR WIRE

(\*\*) WATER CONTENT DEPENDS UPON WATER IN SAND.



(This is a copy of Circular Letter No. 22 from the Office of the Surgeon General, War Department, Washington, D. C., 16 January 1943. Portions referring to use of drugs which have been superseded by newer knowledge have been omitted with omissions indicated by blank spaces.

The use of drugs is presented on pages indicated following this letter).

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WAR DEPARTMENT  
Services of Supply  
Office of the Surgeon General  
Washington

January 16, 1943.

CIRCULAR LETTER NO. 22

Subject: Military malaria control.<sup>1</sup>

1. Introduction. The purpose of this letter is to provide Medical and Sanitary Corps officers with some practical details of measures for the control of malaria under military conditions.

2. Classification of control measures. Military malaria control requires the protection of troops in movement as well as in fixed positions. In each of these two situations both group and individual control measures must be considered. This is the basis for the following classifications.

a. Measures applicable to fixed installations.

(1) Environmental measures.

- (a) Protection against adult mosquitoes.
  - (1) Selecting camp sites.
  - (2) Screening buildings.
  - (3) Spray-killing with pyrethrum extract.
- (b) Control of mosquito larvae.
  - (1) Draining.
  - (2) Filling.
  - (3) Using larvicides.
  - (4) Miscellaneous.

(2) Individual measures.

- (a) Curative treatment.
- (b) Sleeping nets (Mosquito bars).
- (c) Repellents.
- (d) Protective clothing.
- (e) Malaria instruction and discipline.

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<sup>1</sup> It is desired that a copy of this circular letter be placed in the hands of every officer in the Medical Corps, U. S. Army.

b. Measures applicable to field operations.

(1) Individual measures.

- (a) Sleeping nets (mosquito bars).
- (b) Repellents.
- (c) Protective clothing.
- (d) Prophylactic treatment.
- (e) Malaria instruction and discipline.

(2) Environmental measures.

- (a) Protection against adult mosquitoes.
  - 1. Spray-killing with pyrethrum extract.
  - 2. Selecting suitable camp sites.
- (b) Control of mosquito larvae.  
(Should be carried out whenever feasible).

3. Fixed installations. Effective control of malaria in permanent and semi-permanent camps, posts, fields, and stations requires a high degree of cooperation and malaria discipline. Responsibility for malaria control is vested in the commanding officer who depends on Medical and Sanitary Corps officers for surveys, recommendations, and supervision, and Quartermaster and Engineer Corps officers for antimalaria supplies, equipment, and labor, and frequently on the latter corps for prosecution of control measures. Adequate protection in highly endemic areas is not possible without teamwork in planning, accomplishment, and maintenance of the antimalaria measures outlined below.

4. Proper selection of camps. The selection of a suitable camp site is an important antimalaria measure. Malaria is a communicable disease, with mosquitoes acting as the vectors. A native village in which there are numerous infected persons is a health hazard to troops which camp within flight distance of the malaria-carrying mosquito. So, too, a large breeding place of these mosquitoes, if located within easy flying distance of a camp, is a menace. The effective flying range of malaria mosquitoes rarely exceeds a mile in the tropics but is apt to be more than this in temperate zones, or under exceptional conditions of wind or terrain in the tropics. As a general rule, camps should never be located nearer than a mile from malarious villages or important breeding places of malaria mosquitoes. The importance of a breeding place depends not on its appearance or altogether on its size, but on the malaria-carrying capacity of the mosquitoes breeding in it. It is equally important, where feasible, not to allow natives to live on a military reservation, or within a mile of military quarters, for they will provide an active replacement pool of gametocytes.

5. Screening. The following general principles should be observed if screening is to give its full value as an antimalaria measure.

a. Suitable wire of proper mesh should be used. Many types of wire screening are made. Steel wire cloth is most common, either painted or electrogalvanized. The quality of paint and of galvanizing may vary greatly in different lots of wire. Pure copper wire, bronze wire (90 percent copper, 10 percent tin), monel wire (nickel-cobalt alloy), stainless steel, and aluminum have all been used. In most areas a good quality electrogalvanized screen will be satisfactory but in the tropics, especially along the

sea, it is better to use wire of a noncorrosive material, such as bronze or aluminum. A hard drawn wire, 99.8 percent copper, and of heavy grade (having a diameter of 0.015 inch) is suitable. Aluminum wire screening is also good.

The diameters of wire and aperture are important. In general, it is safe to use screening which has apertures larger than 0.0475 inch in diameter. Heavy grade copper wire screening, having 16 meshes to the inch, will have a wire diameter of 0.0150 inch and a mesh diameter of 0.0475 inch, but regular 16-mesh wire screening has an aperture of mesh measuring 0.0512 inch, which will not exclude all malaria vectors and will not keep out Aedes species. Standard 18-mesh wire screening has a wire diameter of 0.0090 to 0.0100 inch and a mesh which is 0.0456 inch in diameter.

b. The screening must be so applied that breakage will be minimal, and that the doors and windows will not facilitate mosquito passage. For instance, screen doors should open outwards, and should be on the windward side of a building, if possible. They should be strongly constructed so that they will not sag or warp. They require springs so that they will close automatically. The places where a foot or hand would naturally be applied to open a screen should be protected with a cross strip of wood or metal. Screen doors should shut against battens, which are strips of wood or metal, to block entry of mosquitoes through the space between door frame and door.

The zinc in certain paints sometimes used on door or window screen frames and the iron in galvanized nails sometimes used to fasten screening to a frame will react with copper screening and cause splitting at the point of contact.

In highly malarious areas it is desirable to have double screen door barriers with a vestibule between.

c. Careful attention must be paid to the closing of all possible apertures not screened, such as cracks and knotholes, spaces where floor or wallboards have separated, openings between flooring and walls, corner openings where joists come together, holes where window shutter prop-sticks extend into a room for easy handling, ventilating pipes and shafts, etc. Holes may be covered with tin shingles or pieces cut from ordinary tin cans. A filler for holes and cracks may be made by boiling shredded paper and flour into a fairly homogenous mass and then adding sand and cement to form a plastic which may be moulded into the holes. This filler is somewhat pliable and will retain its place fairly well. Toilet paper is a suitable tissue for use in making this filler.

d. Not only sleeping quarters but also washrooms, latrines, theatres, post exchanges, and all places for evening recreation should be screened.

e. So far as possible, especially in the tropics, hutments and build-

ings should be so constructed that there is four-way ventilation. The screened buildings must not be so poorly ventilated that occupants are driven out into the open by excessive discomfort. It is wise to provide electric fans or punks where possible in the tropics because they tend to make screening less irksome.

f. Proper routine maintenance of screening is essential, with prompt and effective repair of rents and tears, and discovery and blocking of new cracks and knotholes. In malarious areas the great importance of these apertures for mosquitoes is out of proportion to their seeming insignificance. The soldiers who occupy hutments and barracks should be taught to make all minor repairs. Strict supervision of screening is essential. Under some conditions it may be desirable to assign an enlisted man as mosquito-proofing maintenance orderly whose duties it would be to inspect all screening at regular intervals, making such repairs as are within his capabilities and reporting others to proper authorities.

6. Spray-killing adult mosquitoes. a. General principles. One of the most important measures of malaria control is the spray-killing of adult mosquitoes with pyrethrum extracts. This measure, when used against malaria vectors which rest indoors in the daytime, will (1) immediately destroy a high percentage of infective mosquitoes of an area (2) destroy many potential vectors before they can become infective. The use of pyrethrum sprays will also have a repelling effect, deterring mosquitoes from entering sprayed dwellings or even circumscribed outdoor areas which have been sprayed. Finally, if applied often enough, spray-killing will reduce the density of a mosquito population in an area.

In malaria seasons, it is essential to spray all types of adult mosquito resting places. Whenever possible, such spraying should be extended to native villages within a radius of a mile. Shelters may include not only barracks, houses, hutments, and tents, but also outbuildings, latrines, storehouses, stables, cowsheds, empty boxes and barrels, etc. If troops are operating in hostile areas, local villagers may abandon their homes, leaving behind infective mosquitoes. It may be even more important to spray such villages than the actual quarters of the troops. If the malaria vectors tend to leave houses, tents, and barracks at sunrise, the spraying should be done either before dawn or after sunset. When applied after sunset, the spray will have both a killing and repelling effect.

In military installations in highly malarious areas daily spraying is advisable. Huts at road blocks, sentry huts, huts used by beach patrols, antitank gun emplacements, pill boxes, and similar places when occupied at night may be sprayed advantageously at dusk and again at midnight. It is not necessary, however, to spray native villages oftener than twice a week, since the aim here is to kill fed mosquitoes before they become infective, 9 to 12 days or more after feeding on a gametocyte carrier.

b. Pyrethrum insecticides. Pyrethrum flowers contain certain active

principles which are grouped under the term pyrethrins, and which are extracted by kerosene or other solvents. These pyrethrins kill all species of mosquitoes and certain other insects by destructive action on the central nervous system. The sprays are nontoxic to man and animals (although liberal application of a kerosene-pyrethrum spray to the skin may cause local irritation and inflammation). Pyrethrins rapidly disintegrate by a photochemical catalytic reaction when exposed to sunlight and oxygen. Pyrethrum concentrate and sprays should be kept in tightly stoppered, light-proof containers. Containers should never be left open or in the sun. In sealed containers pyrethrum extracts will maintain their potency for a year or more even in the tropics. If a pyrethrum spray fails within three minutes to kill all the adult mosquitoes with which it comes into contact, it no longer contains the standard amount of pyrethrins and it is not suitable for use.

Three types of pyrethrum products are supplied by the Quartermaster. There is first a 20-1 concentrate, each gallon containing the oleoresins of approximately 20 pounds of flowers, with not less than 75 to 100 grams of total pyrethrins per gallon, or 2 to  $2\frac{1}{2}$  grams per 100 c.c. This 20-1 concentrate in Army practice is diluted with 14 parts of a good quality of water white, preferably odorless, kerosene and may be sprayed from various types of mechanical sprayers, such as are described below:

Secondly, in areas where there may be difficulty in obtaining kerosene for diluting the concentrate, a prepared ready-to-use pyrethrum spray is supplied. This conforms to Class AA rating, as defined in the Department of Commerce Standard Specifications. This rating is based on actual performance killing tests and these AA sprays will contain 150 to 180 mg. of pyrethrins or their equivalent per 100 c.c. With either of the above two sprays, about one-half ounce is required to spray effectively 1,000 cubic feet.

Finally, pyrethrum is supplied in containers holding a mixture of 20-1 concentrate, oil of sesame, and liquid freon, as described below.

c. Freon-pyrethrum aerosol. Pyrethrum may be dispersed from pressure cans or cylinders containing a mixture of 1 percent pyrethrins, 2 percent oil of sesame and 97 percent freon. (Freon 12 is dichlore-difluoromethane). The oil of sesame is a synergist or activator, and enhances the killing power of the pyrethrins. The vapor pressure of the freon produces the necessary spraying pressure, which does not decrease as long as a drop of liquid is present in the closed container. As the freon containing the insecticide is sprayed it forms a fine mist from which the solvent evaporates almost immediately, leaving the pyrethrum and sesame suspended in the air as a cloud of fine droplets called an aerosol. The freon is nontoxic to man and mosquito and it is non-inflammable. It is used simply as an expellant to disperse the pyrethrum and oil of sesame.

The pressure in freon cylinders varies with temperatures. For example,

it is 37 lbs. per sq. in. at 40° F., 84 lbs. at 80° F., 116 lbs. at 100° F., and 205 lbs. at 140° F. Various types of freon-pyrethrum pressure cans and cylinders are available. One pound of freon-pyrethrum mixture is sufficient to spray about 150,000 cubic feet of space when properly used. It is liberated in 12 to 14 minutes of continuous use. To spray a room, hutment, or native dwelling, the can is carried rapidly toward all corners of roof, ceiling, or floor while the spray is allowed to escape. No direct hits on mosquitoes should be attempted, as this wastes spray. About 4 seconds of spraying per 1,0000 cubic feet is usually sufficient in military huts. Somewhat longer spraying for the same cubage is generally required for native huts. It is best to spray under the eaves of a hut before going inside. The freon-pyrethrum spray is so effective that it can be used sparingly and without wastage.

When a mixture of freon and the usual pyrethrum concentrate freezes, a wax is precipitated which may block the tube of the dispenser. The wax goes back into solution after a few days at 70° F. or higher so that this freezing effect is rarely troublesome in actual practice, especially since the dispenser is generally used in warm countries. Within a few months a wax-free concentrate will be available.

d. Hand atomizer or spray guns. Small household-type spray guns are useful for casual spraying of quarters. Where mosquitoes are a nuisance, small spray gun atomizers should be issued at the rate of one per 20 men. They consume relatively greater amounts of insecticide than other types of sprayers but may be used to advantage by the troops themselves for occasional spraying.

e. Paint gun sprayer assemblies. Pyrethrum insecticides can be effectively sprayed through any ordinary paint gun if a source of air pressure of 15 pounds or more is supplied. The source of the pressure may be in tanks pumped by hand or by gasoline or electric motor-driven compressors. Solidified carbon dioxide (dry ice) when available, in suitably constructed pressure tanks is a good expedient.

7. Drainage. It is highly important that fixed installations have suitable antimosquito drainage, integrated with the usual system of drains designed to carry the normal surface water runoff. Drainage should be undertaken early, during the actual construction period. It is desirable to have it well in hand before camp or station is occupied. It is also important to avoid blocking an existing draining system by new construction.

No drainage scheme should be undertaken without careful preliminary surveying and planning. Drainage to control mosquitoes may be accomplished by the use of surface ditches, subsurface drains, vertical drains, pumps, tide gates, and other devices. In every case the aim is to drain an area in such a way that regardless of weather or tide there will be no mosquito breeding. Ditches and drains may be unlined, lined, or rockfilled. The lining may be precast sectional inverts, concrete or stone slabs, rip rap, or other material. For subsurface drains many materials have been

utilized, as, for example, baked clay or concrete tile, rock, bamboo, and brush.

Drains should be as few as will accomplish the purpose. Drainage requires that careful levels be run and that well-thought-out plans be made as to the relation of a drainage system to existing water supplies, to other sanitary improvements, to disposal of sewage, and to maximum amount of water to be carried in the rainiest season. In some areas, where the vector breeds in seepage or in slowly running water, open ditches may cause a greater malaria nuisance than the swamp they drain.

Where a satisfactory outlet is not available, land may sometime be graded so that surface water accumulates in one or more pools which can be periodically treated with larvicides.

Ditch maintenance is a very important item for which adequate provision must be made.

8. Filling. It is usually necessary to do a considerable amount of filling on the site of a new permanent installation. Frequently, there will be man-made depressions under buildings, beside roadways, and in other places where earth has been borrowed for various purposes. When large hydraulic fills are contemplated for construction purposes, it is necessary to make adequate plans to avoid blocking natural drainage and to control with larvicides the breeding places, such as pools and cracks, created by filling operations.

9. Larvicidal oiling. a. Discussion. Suitable oils, properly applied, will kill the aquatic stages of all species of mosquitoes and will also destroy sheltering vegetation at the edges of breeding places. The chief killing factor is a toxic action following contact with the tracheal cells of larvae and pupae. Consequently, the best larvicidal oils are those which penetrate most quickly, and with the greatest toxic effort, into the spiracles and thence into the tracheae of larvae and pupae. What is required is a cheap but toxic oil or mixture of oils of suitable toxicity and viscosity which, when sprayed on the surface of the water, spreads well in a uniform, persistent, and stable film. The best larvicidal oils will kill in less than 30 minutes under these conditions.

Kerosene or gasoline may be used as larvicides and will give a good kill but they form transitory films, are expensive and may constitute a fire hazard. Occasionally it may be desirable to kill all larvae in a well by a film of gasoline, which soon evaporates, leaving no taste in the water. Leaded gasoline should not be used in wells the water of which is used for drinking.

Waste motor oils are not highly toxic to larvae, as they are relatively nonvolatile. But they are sometimes used effectively when applied as a fairly thick film. Better results are possible when wasted motor oil is

mixed with kerosene, generally in the proportion of 1 to 3, respectively, adding if possible about 2 percent of castor oil. However, the amount of kerosene to be added will have to be determined by experiment.

Diesel oil No. 2, as supplied by the Corps of Engineers, is preferable to a mixture of waste oil and kerosene.

The ideal specifications for a larvicidal oil are the following:

Specific gravity 20/4	0.83 - 0.86
Viscosity (Saybolt Universal at 100° F.).	31 - 43
Initial boiling point	297°- 414° F. (165°-230° C.).
Final boiling point	Max. 800° F. (426.70° C.).
Spreading coefficient	Min. 17.0

b. Application of oil. General. Oil may be effectively applied to small collections of water by means of an oil-soaked broom, and oil mop, or oil-soaked waste tied to a stick. An ordinary waterpot may be used to pour oil on small collections of water.

c. Sprayers. The knapsack sprayer consists of oil container, hand pump, and spray nozzle, and is carried and operated by one man. The ordinary sprayer has a capacity of 4 to 5 gallons and a spraying range of about 25 feet. The knapsack sprayer is a practical and economical apparatus for applying oil to ditches, small ponds, or other collections of water which can be reached by the spray. Large power sprayers may be employed to oil extensive areas such as the borders of lakes, or, in some instances, swampy places. Such sprayers usually consist of a barrel or tank container and a motor activated pump mounted on a vehicle or boat. Airplanes may be equipped to oil extensive breeding areas.

d. Continuous oilers. Where oil will be dispersed by currents, as in streams or ditches, it may be better to attempt constant application of oil. There are at least three types of continuous oilers, none of which is entirely satisfactory. It must be pointed out that continuous oilers require a good deal of attention, generally use more oil than would be dispensed for the same area from a knapsack sprayer, and should not be used unless other methods are found impractical.

Drip oilers consist of a tin or drum of about 5 gallons capacity, placed on supports over a stream or ditch so that oil will drip on the water surface. Size of hole will govern amount of oil dropping from the container. In home-made containers a nail hole may be used, with a nail left loosely in the hole. It may be necessary to use some string to form a washer around the nail head. The can should be several feet higher than the stream surface so that oil will spread quickly when drops strike the water. The rate of flow required to furnish a satisfactory film depends on circumstances. Generally, an average flow of from 10 to 20 drops per minute will suffice for each foot of width of water in the stream.

Submerged oilers are containers having two small openings. They are designed so that when sunk to the bottom of a stream or pond their oil will escape through one opening and be replaced by water which enters through the other. These cans have the disadvantage that they are difficult to adjust so that oil will flow properly, as the openings are easily clogged.

Oil may be applied continuously by means of a weighted submerged bag of oil-soaked sawdust. Or oil-soaked sawdust may be scattered over the surface of a breeding place.

Oil booms may be used to combat larval drift, which is sometimes troublesome when there is intense breeding in a stream above the area being controlled. These booms may be constructed of locally obtained materials and placed across a water channel, supported by stakes. The booms are so built that, while holding back larvae, the flow of water is not obstructed. The larvae are killed by oil-soaked sawdust or chaff thrown on the water surface above the boom.

c. Amounts of oil required. Using diesel oil No. 2, about 9 gallons are required per acre of water surface for complete coverage with a uniform stable oil film. With an ordinary knapsack sprayer of the Panama type one laborer can oil about five acres of breeding area per day, if the terrain is not difficult. In usual practice the amount of oil necessary to produce a uniform film may vary from 10 to 20 gallons per acre. The amount of floatage and vegetation will make a considerable difference. It is usually necessary to spread oil once a week.

10. Spreading Paris green on mosquito breeding places. a. Nature of Paris green. Paris green (an aceto-arsenite of copper) is a micro-crystalline powder. It should be emerald green, with at least 50 percent arsenious oxide. Its solubility should not exceed 3 percent. Paris green should pass a 300 to 325 mesh sieve. The latter has a mesh opening measuring 0.043 mm. As mixed, ready for distribution in the field, Paris green should kill all 2nd, 3rd, and 4th stage larvae in a laboratory jar in two hours. It has a specific gravity greater than that of water, but certain types float longer than others. The longer it floats the better it is as an anopheline larvicide.

The usefulness of Paris green depends on the fact that it is a gut poison for larvae. Larvae feeding at the surface will ingest Paris green particles. This is especially true of Anopheles larvae, but when Paris green floats long enough in still water it will be ingested by Aedes and some Culex larvae, which feed some of the time at the surface. Sometimes when Paris green is mixed with wet sand and taken to the bottom, it will be ingested by bottom feeding larvae. Paris green, however, has its greatest usefulness against Anopheles larvae.

Paris green, after sinking is usually volatilized by various moulds in the water. As ordinarily used in mosquito control procedure, it is harmless to man, animals, fishes, and bankside or floating vegetation.

Paris green does not repel ovipositing mosquitoes and in this respect is inferior to oil.

b. Dust dilutions of Paris green. When dust mixtures are used, it is advisable to prepare a diluent which will pass a 30-mesh screen, or finer. This diluent may be road dust, powdered charcoal, slaked lime, powdered soapstone, or some other dust. The diluent must be well mixed with Paris green, the required dilution varying from 1 to 5 percent by volume for hand operated blowers to 25 percent or more for dusting from airplanes. When lime is used, Paris green may be added in the proportion of 10 percent by weight. This is equivalent to about 5 percent volume.

However distributed, it is essential to test the efficiency of Paris green in the field from time to time. This may be done by ascertaining the kill in petri dishes containing larvae and placed at strategic points in the dusted area, or by dipping for larvae 10 to 24 hours after application of the Paris green and noting the survivals.

c. Types of dust distributors. Paris green mixtures can be applied to ponds, lakes, and larger streams by putting the dust into the air from the windward side so that it will form a cloud and be carried along over the water. The large hand or motor operated dust blowers ordinarily employed for dusting trees in horticultural work may be used to throw the larvicidal dust into the air. These may be knapsack, rotary, or some other type. For large bodies of water a slowly settling dust cloud carried along by a light wind will give best results. In the case of small bodies of water where vegetation is heavy or where ditches and streams are too narrow to be dusted by the cloud method, handfuls of dust mixture should be thrown directly on or into the vegetation or onto the surface of the water. There are also automatic distributors for small canals or streams. Airplanes may be used to apply the dust over large areas, such as extensive swamps. Generally, the best results will be obtained on a sunny day after the dew has evaporated from vegetation.

It should be remembered that, when special distributors are not available, Paris green mixed with wet or dry sand, gravel or small pebbles and thrown by hand is often quite effective.

The amount of larvicide required to treat an area of given size will vary according to the amount of vegetation present. When the surface of the water is clear, about one-half ounce of Paris green, mixed with 99 parts of dust by volume, is sufficient for 1,000 square feet of water surface. Greater quantities of Paris green must be used where there is vegetation, such as grass or reeds. The percentage of Paris green in such places should be from 2 to 5 percent by volume, as determined by experiment.

One man can usually prepare and spread Paris green mixture along about  $1\frac{1}{4}$  miles of bank in one day depending, of course, on local conditions.

Since Paris green will kill 2nd, 3rd, and 4th stage larvae within a

few hours it need be applied only at such intervals as are necessary to prevent the development of the remaining 1st stage larvae or of new broods of larvae. Under average conditions Paris green should be applied at intervals of from 5 to 7 days in warm weather. Dusting should be repeated without delay whenever examination of the water reveals the presence of 4th stage larvae.

d. Dustless application of Paris green. The need for preparing and transporting dust diluents is obviated by using a dustless method with the following stock suspension:

Kerosene oil	1 pint
Paris green	1/2 pint
Egg albumen (dry powdered)	1/4 teaspoonful

(If no powdered egg albumen is available an albumen solution can be prepared by using 4 to 6 egg whites in a pint of water. Use one-fourth of a pint of this solution to one pint of kerosene. The egg albumen is not absolutely essential but it tends to make the Paris green more evenly distributed in the final spray). The ingredients are put into a bottle in the order named and thoroughly shaken. This stock suspension, again thoroughly shaken, may be put into vials, each holding 6 teaspoonfuls (about 27 c.c.). These stoppered vials may be taken to the field by the spraying laborer in a specially made belt.

To prepare the final spray one vial, or 25 to 27 c.c. of stock suspension, is mixed with 5 quarts of water. It reduces the labor of transportation, if the mixing is done at the breeding place to be sprayed. The water should be strained through a sieve and the mixing may be done in the usual knapsack sprayer, which generally has about a 4-gallon capacity. By using only 5 quarts of final spray at one time in this sprayer it is possible by frequent agitation to keep the spray well mixed while it is being applied. About 500 square feet can be sprayed with 5 quarts of this spray.

About 2 teaspoonfuls of castor oil in 5 quarts of spray will add to the effectiveness of this dustless Paris green larvicide.

When it is not possible to prepare the above stock suspension, fairly good results may be had with a simple mixture of 5 teaspoonfuls (about 22.5 c.c.) of Paris green in 5 quarts of water, using an ordinary knapsack sprayer. Almost continuous agitation of the sprayer while spraying is advisable. Some 500 square feet of breeding area can be sprayed with 5 quarts of this larvicide.

11. Care of equipment. Equipment for spreading oil or Paris green larvicides requires careful maintenance. It should be overhauled and thoroughly cleaned at reasonable intervals. Full sets of replacement parts should be stocked.

12. Phenothiazine. It has been shown that phenothiazine is an effective substitute for Paris green. If available, in the absence of Paris green, it may be used. It is less stable than Paris green but more floatable.

13. Miscellaneous measures to deal with breeding places. In some areas, effective larva control may be had by shading a stream or canal, using some rapidly growing plant which will give dense cover.

In other areas, stream breeding anopheline vectors may be controlled by periodically sluicing the stream. A small dam is built to impound water which can be released by the hand or mechanical removal of a barrier, or by use of automatic siphons. Where irrigation water in rice fields or canals is at fault, effective mosquito control is sometimes possible by intermitting the supply of water so that the fields become just dry enough each week to remove the surface film of moisture without drying the roots of the plants. Fluctuation of water level in ponds and reservoirs is sometimes very effective in controlling mosquito breeding.

All wet cultivation, such as sugar cane or rice, within two miles of any fixed Army installation, should be eliminated if feasible.

When larvicides such as oil and Paris green are not available, control of breeding is sometimes possible by tightly packing a ditch or small pool with fresh green-cut vegetation. Unless tightly packed and renewed as required, the rotting vegetation may create conditions leading to increased culicine breeding.

In some areas routine attention must be paid to artificial breeding places such as wells, cisterns, roof gutters, and various types of household water containers, tin cans, and coconut shells. These may be screened, emptied, oiled or destroyed, as indicated.

In wells and in pools without much vegetation some help in larva control may be had by using larva-eating fishes, such as Gambusia affinis.

14. (Omitted).

15. Malaria Control in the field. "Fighting" malaria control, or the prophylaxis of malaria among troops in combat or maneuver areas in the field, is one of the most difficult of all malaria control problems. It requires constant attention to the application of individual protective measures against mosquitoes and to the use of suppressive antimalarial drugs. There must be the greatest accent on personal prophylaxis. The soldier must himself apply measures which will protect him from malaria. Under combat conditions the need for freedom from malaria is greatest, yet under these conditions so is the risk of infection. Therefore, it is highly important to apply effectively such measures as suppressive treat-

ment and the use of sleeping nets, repellents, sprays, and protective clothing.

16. Use of sleeping nets. Nets to protect sleeping individuals are useful in preventing malaria but they must be properly employed and properly maintained. They must be so adjusted and used that mosquitoes cannot feed through the mesh because the net touches the individual. The lower edge must be so tucked in that no opening is available for mosquitoes to enter. Overhead frames should be provided for bed and cot nets. These should not have sharp points which will catch and tear the netting. Nets used in small tents should be suspended from and conform to the shape of the interior of the tent. Shelter tent nets should not be used over the outside of the tent but hung inside. Nets should be folded up by day. When the net is entered at night, the interior should be inspected for stray mosquitoes.

It is highly important that nets be available for use from the first night spent in malarious areas. There are places in the tropics where a single night of exposure to mosquito bites may result in a 20 percent or greater infection rate among the exposed troops. Nets should therefore be carried as items of personal equipment by all personnel proceeding to malarious zones. Even in the forward areas it is highly important to utilize this protective measure whenever feasible.

The best material for nets is a stiff bobbinet. Such materials as cheesecloth, tobacco cloth and butter cloth, are not suitable because they almost completely prevent proper ventilation. The size of the holes should not exceed 0.0500 inch in any dimension to exclude all species of mosquitoes. To exclude sandflies the largest permissible diameter is 0.0334 inch. The top as well as sides should be of netting to allow for a better circulation of air. Strong reinforcing at corners is necessary and repairs should be prompt and complete. Adhesive tape, sewing, or patching may be used to repair rents in the netting. Frequent inspection is required.

17. Use of chemical repellents. Various essential oils and synthetic products have been used, as creams or lotions applied to the skin, to repel mosquitoes. Most mosquito repellents have had one or both of two major defects: (1) Very transitory or weak effect, and (2) Risk of toxic poisoning by absorption through skin, especially when the repellent must be used liberally during extended periods of time. For example, diethyleneglycol is a mosquito repellent but is reported to be toxic when absorbed through the skin and apt to damage kidney and liver tissue if used freely for a considerable time.

There are three good repellents (612, indalone, and dimethylphthalate) which are being issued by the Quartermaster. Of these, 612 will give good protection against mosquitoes for about four hours after liberal application even under sweating conditions. Indalone will do about as well, except under sweating conditions when it should be renewed half-hourly. Dimethylphthalate is slightly less effective than 612, but more effective than indalone. All are better than any repellents available heretofore.

Certainly very much greater use than ever before should be made of these repellents when troops are in forward areas. All exposed skin surface should be covered with the repellent. It is also important to apply the repellent to the clothes where they are so thin or so taut that mosquitoes can feed through the cloth as, for example, over the shoulders and seat. Repellents should not be applied to the lips, nor allowed to reach the eyes.

18. Use of protective clothing. Men on guard duty or forced to remain out after dusk where malaria-carrying mosquitoes are common, may wear approved head nets and gloves as supplementary protective measures. Head nets have certain disadvantages. They impair vision and add to discomfort in the tropics. Vision is least impaired by black nets. Shirt sleeves should be kept down after sunset. Slacks, not shorts, should be worn. By keeping the trouser legs encased in leggings, or tucked into high boots, mosquitoes are kept from biting the ankles. Mosquito boots of canvas, suede, or goatskin may be useful if long enough to protect the leg nearly to the knee. Such light boots are not suitable for marching, which requires service shoes with leggings.

The efficiency of protective clothing varies directly with the amount of inspection and discipline which can be applied.

19. Pyrethrum sprays. It seems likely that pyrethrum sprays, especially the freon-pyrethrum aerosol, will be found increasingly useful in the control of malaria in the field away from fixed installations. The repellent effect of pyrethrum is so marked that, although adequate tests have not been reported, it nevertheless seems advisable to suggest that in highly malarious areas much greater use be made of pyrethrum sprays in temporary encampments and even in front line positions. Shelter tents and jungle shelters should be sprayed at dusk, and any native huts or outbuildings in the vicinity should also be sprayed, when feasible. There is evidence that the spraying of the outer garments will help materially to repel mosquitoes.

20. Instruction of personnel. The instruction of all ranks in regard to malaria and malaria control is important, and it should begin in the continental training bases, even if these are not malarious. The nature of malaria, cycle of development of plasmodium, role of Anopheles mosquitoes, and the fact that the disease can be prevented only by special measures, should be explained briefly in simple terms. Full use should be made of moving pictures and of locally prepared talks, bulletins, and directives. An important point to be stressed is that malaria cannot be prevented by routine camp sanitation or personal hygiene. Specific control measures are essential. In permanent and semi-permanent posts and camps a high degree of protection against malaria is afforded by employing measures outlined above. In forward or combat zones and while maneuvering or holding defense positions, in the field away from fixed installations, especially in tropical countries, the most feasible malaria control measures are those for which the individual soldier himself must be responsible to a very large extent.

21. Drug prophylaxis, or suppressive treatment. a. General. Drug prophylaxis or chemoprophylaxis against malaria was attempted long before the ediology of the disease was known. It is now clear that (1) true or so-called causal prophylaxis, i.e., the actual prevention of infection, is not certain by the use of any known drug; (2) sporozoites do not appear to be destroyed in the body by drugs; (3) malaria probably cannot be eradicated from a community by the use of drugs.

(Omission).

e. Discussion of chemoprophylaxis. As already stated, there is no drug which in safe doses will prevent mosquito-borne infection with malaria. But quinine and atabrine, in small doses, are almost equally useful in suppressing the appearance of clinical symptoms after infection. Such suppressive treatment has great emergency value in that it will enable malaria infected troops to maneuver and fight actively in spite of the infection, which otherwise would incapacitate them. The usual clinical rigors and pyrexia are suppressed and do not appear until prophylactic medication has been stopped. In dealing with large numbers of men, the terminal point of prophylactic drug administration should be staggered to avoid overcrowding the hospital with relapsing cases, which may occur to the extent of 50 percent or more, whether quinine or atabrine has been taken. Except for this possible need to stagger the terminal point, there is no logical reason to continue the prophylactic drug after a man has returned to a protected area. He should then be observed for indications of malaria and given a therapeutic treatment if required. During this period of observation repeated thick blood smears should be examined, if possible.

(Omission).

22. Miscellaneous. a. Nocturnal visits to unprotected places. In some areas a high percentage of all malaria infections is acquired while soldiers are visiting unprotected towns or villages after dark. In such areas it is advisable to consider a change in the usual leave hours whereby passes are issued which will require the soldier to return to his protected camp by nightfall.

b. Malaria discipline. Malaria discipline is defined as a state of orderly and effective conduct or action on the part of soldiers in respect to malaria control. It implies ability and readiness to practice malaria control, as outlined above, particularly the individual measures. When there is malaria discipline in a company, screens and bed nets are in good repair and are properly used, protective clothing and repellents are employed to the fullest practical extent, suppressive treatment is faithfully taken as ordered, and men do not expose themselves heedlessly to mosquito bites. Good malaria discipline reduces to a minimum such dangerous practices as loitering, fishing, or swimming after dusk in malarious

places.

Malaria discipline is developed by careful indoctrination of officers and enlisted men, and it presupposes constant supervision by those in command.

Malaria control is never automatic. In highly infested areas it requires of the Medical and Sanitary Corps officers concerned unremitting attention to small details. Success is possible only when there is a high degree of cooperation among all ranks and all branches in outwitting the Anopheles mosquito.

For The Surgeon General:

Signed:

John A. Rogers,  
Colonel, Medical Corps,  
Executive Officer.

Distribution:

Each officer of the  
Medical Corps, U. S. Army

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NOTE: Any possible discrepancy between the mimeographed copy made by the Army School of Malariology and the original mimeographed letter from the Surgeon General's Office would exist through inadvertence and would be the responsibility of the present Senior Author and Editor.

## FOREWORD

### USE OF DRUGS AGAINST MALARIA

Detail of medication for suppressive and curative treatment of malaria is necessarily recorded with caution and hesitation. In these days of rapidly expanding investigation and experimentation there is a concomitant and associated growth of knowledge founded on fact. The empirically accepted "truth" of today may be the recognized scientifically proven error of tomorrow.

The discussion of medication which follows is extracted from TB Med Nos. 65 and 75, dated July 3rd and 10th, 1944, and revalidated February 24, 1945.

It is herewith included to replace those portions of the preceding Surgeon General's Circular Letter No. 22 which have been in instances superseded by more recent knowledge.

New drugs are now in process of experimental trial and these in turn may later supersede present accepted procedures.

## DRUG SUPPRESSION OF MALARIA

Current Army policies on the drug suppression of malaria are outlined in War Department TB Med 65, entitled "Drug Suppressive Treatment of Malaria", dated 3 July 1944. The following paragraphs summarize the essential features:

1. No drug has been discovered to date which is known to be a causal prophylactic or one which will prevent infection. Two drugs are available, however, which will suppress the clinical manifestations of the disease if administered in sufficient dosage and with scrupulous regularity. Of these two medications, atabrine and quinine, the former is the drug of choice not only from the standpoint of availability but also from the standpoint of toleration, preference by troops, and effectiveness. Military experience has demonstrated conclusively that when the taking of atabrine is adequately enforced, non-effectiveness from malaria can be kept at a satisfactorily low level in combat troops.

2. Experimental studies on plasma concentration of atabrine during suppressive treatment have shown that one half the maximum level is attained in one week increasing thereafter at rate of 50% per week. The delay, which is incident to tissue saturation, indicates that suppressive therapy should be initiated 1 or 2 weeks prior to exposure where possible. A program of suppressive therapy in a unit moving into a malarious area, however, should be initiated only when ordered by proper authority.

### 3. Administration of Atabrine.

a. The recommended dosage is 1 tablet (0.1 Gm) daily at the morning or evening meal, total 0.7 grams per week. It should be administered by roster to both officers and enlisted with a competent non-commissioned officer witnessing the actual swallowing of the drug by each individual. It is recommended that all individuals failing to take the drug be required to report and take sufficient dosage to equal the amount missed; also, men on detached duty, should be given sufficient drug for the period they are to be away with specific directions for taking it.

b. During critical periods of great military urgency such as may occur in actual combat where a high malaria incidence would present a hazard to the mission involved, increased or "loading" doses may be employed at least 2 weeks prior to the projected operation in troops who have become adjusted to the smaller dose schedule. "Loading" may be accomplished by increasing the daily dosage to two tablets daily, (0.2 Gm), or to 0.5 gram doses at intervals of 3 or 4 days. The increased dosage schedules should be discontinued promptly when the critical period is over.

c. Where a considerable number of "break throughs" of clinical malaria occur, particularly following strenuous combat duty, the malaria rate

may be reduced by giving 3 tablets daily after meals and under medical supervision for a period of 3 to 5 days following which the routine schedule of 1 tablet per day should be resumed.

#### 4. Reactions.

a. A yellowish discoloration of the skin which is not a toxic reaction will occur with continued use of atabrine and will disappear within a few weeks after the drug is discontinued.

b. A small percentage of individuals will experience untoward reactions in the form of nausea, vomiting and occasionally abdominal cramps and diarrhea. These symptoms are never serious and almost invariably disappear with continued use of the drug. The extent and incidence of this reaction is unpredictable but reactions may be diminished through the use of  $\frac{1}{4}$  tablet, (0.05 Gm.), as the initial dose. Experience to date gives no evidence of toxicity from long continued use of atabrine.

5. Less than 1% individuals will be found to show a true intolerance to atabrine. In such cases quinine in dosages of 10 grains daily should be employed.

6. In highly malarious areas clinical symptoms during suppressive treatment may occur in a certain percentage of cases particularly when under the stress of combat. Although the majority of such instances represent failures to take the drug regularly, a small number of cases may occur despite good atabrine discipline. In such instances patients with clinical symptoms should be given a full course of treatment following which, suppressive treatment should be resumed if still indicated.

7. There is evidence to indicate that if atabrine in suppressive doses is continued for a sufficiently long period, infections with P. falciparum are cured. It is therefore desirable that suppressive medication be administered for a period of at least four weeks after withdrawal from a malarious area. To this end War Department Circ. 449, dated 25 November 1944, prescribes that individuals returning to the United States under suppressive treatment for malaria be continued under such therapy for a period of 28 days.

a. In units heavily seeded with malaria, suppressive therapy should not be discontinued if relapses will be so numerous as to interfere with training and rehabilitation.

b. In large units where the incidence of infection is unknown, suppressive therapy should be discontinued only in a small representative sample of men in order to estimate the amount of malaria in the entire unit.

c. When suppressive therapy is discontinued in a large force, this may be staggered to avoid over-taxing hospital facilities.

8. It should be emphasized that the administration of suppressive medication is a command function and every effort should be exerted to maintain a high standard of atabrine discipline. It is likewise essential to emphasize to troops that suppressive medication is not curative and that where suppressive therapy is necessary even greater attention to anti-mosquito measures is indicated.

## CURATIVE TREATMENT OF MALARIA

Current Army policies concerning the curative treatment of malaria are outlined in War Department TB Med 72, entitled "Treatment of Clinical Malaria and Malarial Parasitemia", dated 10 July 1944. The following paragraphs summarize the essential features:

1. Uncomplicated malaria and parasitemia without symptoms (patient able to retain oral medication).

a. Atabrine. The drug of choice for routine therapy.

(1) Atabrine dihydrochloride 0.2 Gm (3 grains) and Sodium bicarbonate 1 Gm (15 grains) with 300 cc of water, sweetened tea, or fruit juice every 6 hours for 5 doses.

(2) The above to be followed by atabrine dihydrochloride 0.1 Gm ( $1\frac{1}{2}$  grains) three times daily after meals for 6 days (total dosage 2.8 Gms in 7 days).

b. Quinine. Restricted to instances where atabrine is not available, where there is serious intolerance to atabrine or in selected cases where repeated relapses have occurred following use of atabrine and a change of drug is considered desirable.

(1) Quinine Sulfate 1 Gm (15 grains) 3 times daily after meals for 2 days.

(2) Followed by 0.6 Gm (10 grains) 3 times daily after meals for 5 days (total 16 Gm in 7 days).

c. Totaquine. Limited to selected cases. Not recommended for treatment of P. falciparum infections or for use in forward areas.

(1) Totaquine USP 1 Gm (15 grains) three times a day after meals for 3 days.

(2) Followed by 0.6 Gm (10 grains) three times daily after meals for 5 days (total 16 Gms in 7 days).

d. Plasmochin. Not to be used routinely. May be used under strict medical supervision, where gametocytes persist following routine treatment. Plasmochin must never be administered in combination with atabrine. Patient must be kept in bed during treatment. Discontinue at once if toxic symptoms appear.

(1) Plasmochin naphthoate 0.02 Gm ( $1/3$  grain) with Sodium bicarbonate 1 Gm (15 grains) by mouth 3 times daily after meals for 4 days.

Note: 0.02 Gm plasmochin naphthoate is equivalent to 0.01 Gm plasmochin hydrochloride.

(2) Liberal administration of fluid and sugar during and several days after administration.

2. Malaria with persistent vomiting, coma, impending coma, a high density of P. falciparum parasites in blood stream (5% or more red blood cells infected) or in the presence of serious complicating diseases such as dysentery, pneumonia, meningitis or grave injuries.

a. Atabrine.

(1) Atabrine dihydrochloride 0.2 Gm (3 grains) in 5 cc sterile distilled water injected intramuscularly into each buttock (total 0.4 Gm or 6 grains).

(2) Followed by 0.2 Gm (3 grains) intramuscularly every 6 to 8 hours until oral medication can be instituted.

(3) As soon as oral medication can be retained atabrine should be given by mouth in sufficient dosage to bring the total for the first 48 hours to 1.3 Gms followed by 0.1 Gm (1½ grains) 3 times daily after meals for 5 days. (Total 2.8 Gms in 7 days).

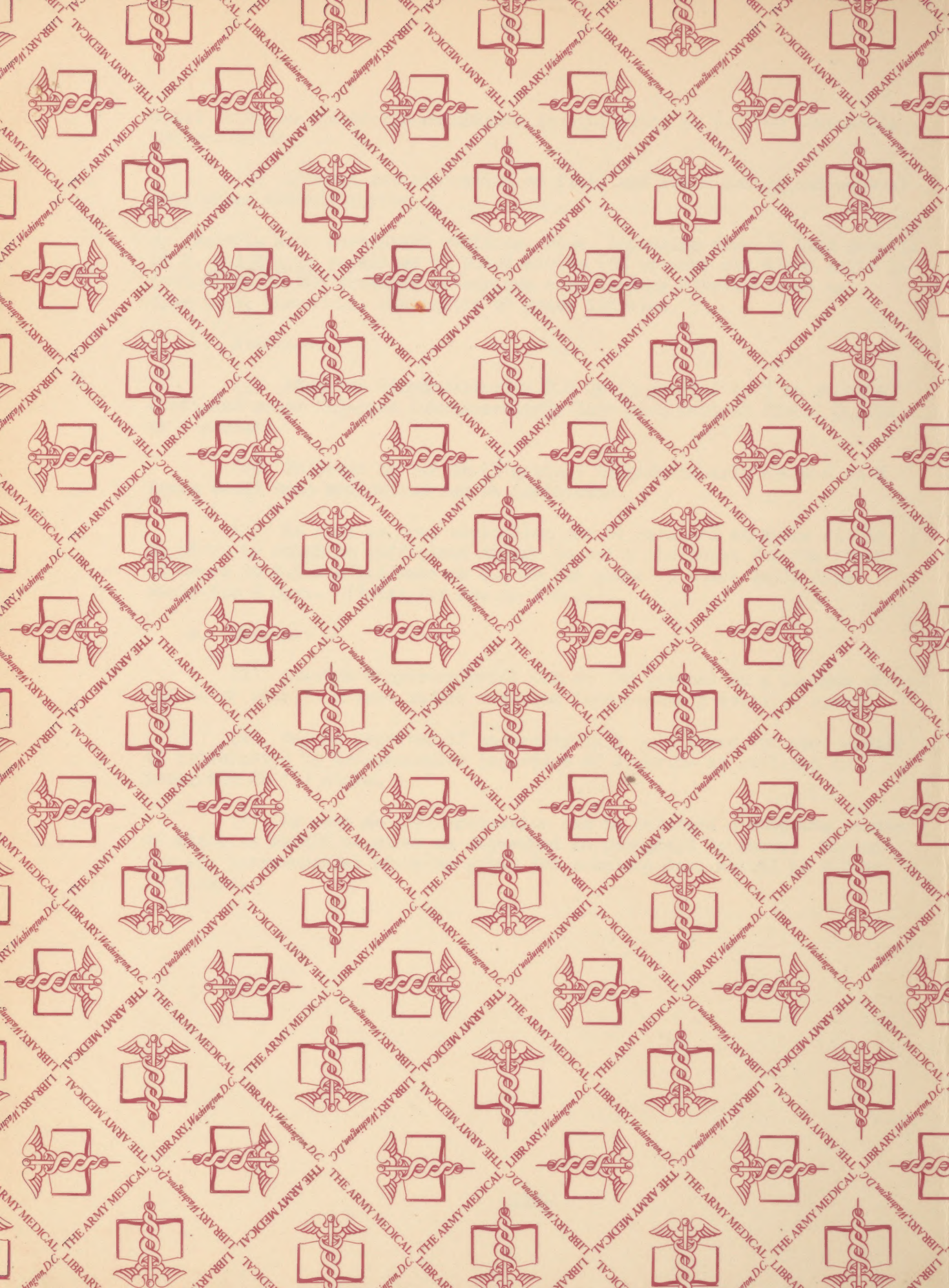
b. Quinine.

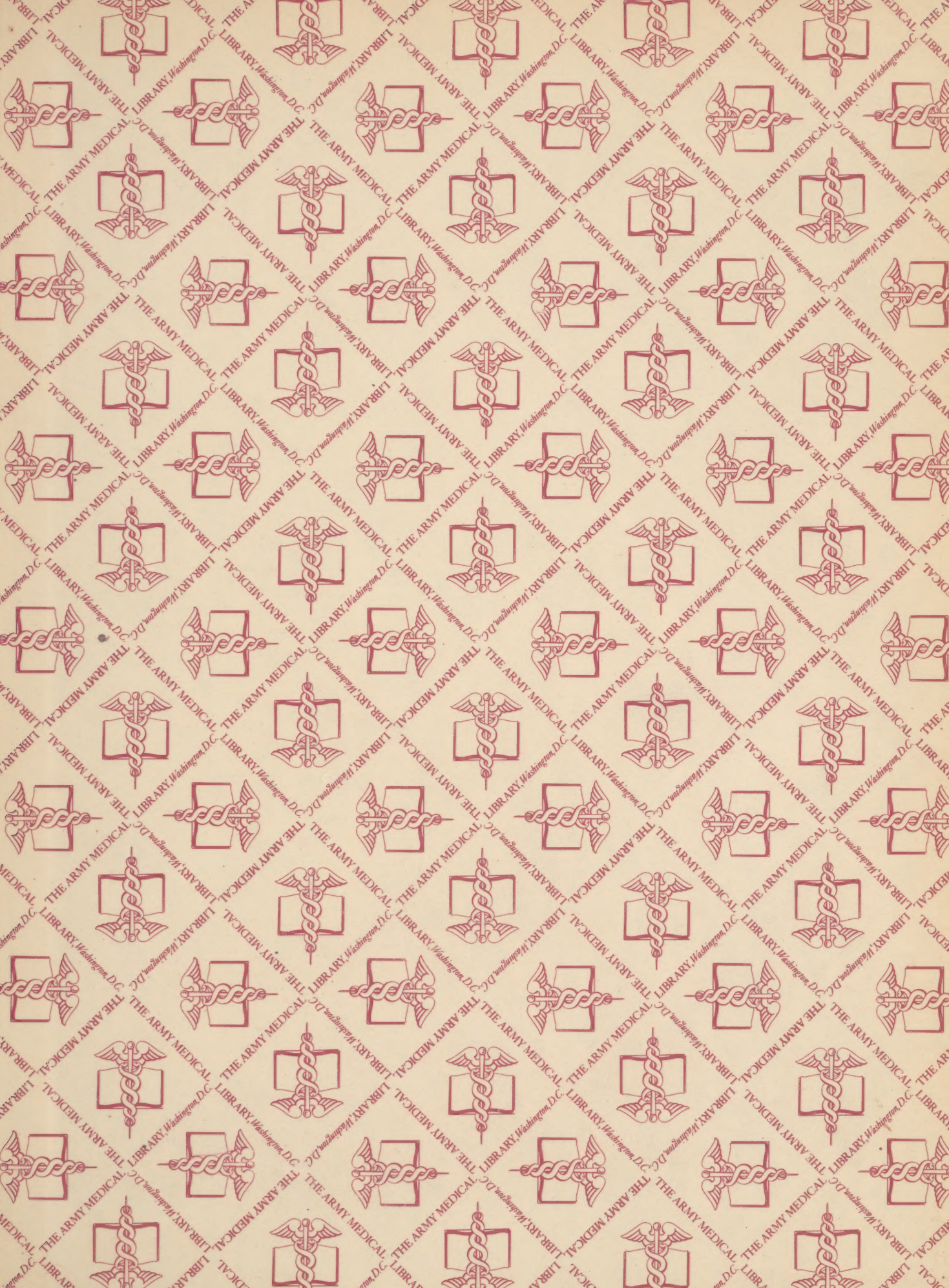
(1) Quinine dihydrochloride 0.6 Gm (10 grains) in a minimum of 200 cc of sterile normal saline injected intravenously avoiding speed.

(2) May be repeated if necessary in 3 to 4 hours but need should be anticipated by intramuscular injection of atabrine as outlined in paragraph 2a (1) above.

(3) As soon as oral medication can be retained a complete course of atabrine should be given as described in paragraph 1a above. Including any intramuscular atabrine given the complete dose should be 2.8 Gm in 7 days.







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